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EFFECT OF COMPRESSION ON MECHANICAL PROPERTIES OF READY-TO-EAT (RTE) SPINACH VARIETIES

SITI HAJAR ARIFFIN[1,2,3*], KONSTANTINOS GKATZIONIS[1,4] and SERAFIM BAKALIS[1]

[1]*School of Chemical Engineering, University of Birmingham, B15 2TT, Edgbaston, Birmingham, United Kingdom*
[2]*Department of Process and Food Engineering, Faculty of Engineering, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia*
[3]*Laboratory of Halal Services, Halal Products Research Institute, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia*
[4]*Department of Food Science and Nutrition, School of Environment, University of the Aegean, Metropolite Ioakeim 2, 81400, Myrina, Lemnos, Greece*
[*]*E-mail: hajarariffin@upm.edu.my*

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ABSTRACT

Compression-induced injury is frequently encountered during handling, packaging, transporting, and storage. In this study, compression test was performed using a mechanical tester, Universal Testing Machine Z030 (Zwick/Roell, Germany). Spinach varieties (Teen, Organic, Salad, and Baby) were used to study the response of spinach varieties towards stress. For single loading/unloading compression, maximum work (MaxW) and area under the curve (AUC) required to compress Organic spinach were found to be the highest followed by Teen and Salad spinach. The MaxW and AUC were found to be decreasing after storage which showed that the total work generated to compress the leaves was reduced due to texture degradation of the product after storage. For multiple loading/unloading compression, as the number of compressions increased, the MaxW decreased. Similar trend was observed at day 6. Apart from that, the MaxW for all the three spinach types were found to be similar at the 5th compression. This shows that regardless of the spinach types, they reached maximum resistance towards stress after the 5th consecutive compression. Under 200 N compression, leaves with stem required higher energy to compress compared to leaves without stem. However, for leaves compressed under 50 N and 100 N, the difference was only noticed on the 1st compression. The MaxW was found to be similar for leaves with stem and without stem starting from the 2nd compression till the 5th compression. The irregular and larger cell size of Organic spinach as compared to round-shaped and smaller cell size of Teen and Salad spinach may contribute to the ability of the Organic spinach tissue to have higher resistance towards mechanical stress during compression.

Key words: *Spinacia oleracea*, spinach, ready-to-eat (RTE) vegetables, mechanical property, loading/unloading compression

INTRODUCTION

Ready-to-eat (RTE) or also known as minimally processed vegetables are defined as fresh vegetables that have undergone minimal processes to increase their functionality while maintaining their fresh-like properties (Dinnella *et al.*, 2014; Ragaert *et al.*, 2004). Food industries have invented various methods and treatments in order to maintain and improve the shelf life of the RTE vegetables. However, uncontrolled or excessive treatments caused accidental damage which then leads to quality and shelf life reduction of the products. Up to date, the industry has been monitoring the quality of the fresh product varieties before they are ready to be marketed. List of criteria is made to ensure the product that passes the quality inspection to know whether it is good enough for the consumer. However, the criteria on choosing the best product are made before the product undergoes the processing steps. Question arises as does the product chosen as the best variety before processing remains the best type after processing? The best variety is not only the one that exhibits good qualities before processing but also the one that is best in resisting stress even after undergone the processes. The

* To whom correspondence should be addressed.

industry needs to understand and should be able to correlate the impact of mechanical injury that caused during processing as well as which types of varieties is the best in resisting the stress. Selection of raw material, effect of processing and storage treatment need to be studied as a whole rather than individual assessment.

There have been extensive works reported on the mechanical properties of fruits and leafy vegetables; however, there is still lack of information to determine the mechanical behaviour of the leafy vegetables in bulk, as it would be retailed (i.e. in packaging bags). Most mechanical properties reported in the literatures were investigated based on individual sample by performing mechanical tests such as tensile test, compression test, penetration test, cutting, tearing, puncturing and shearing (Ávila *et al.*, 2007; Desmet *et al.*, 2004a; 2004b; Golmohammadi, 2013; More *et al.*, 2014; Nabil *et al.*, 2012; Tang *et al.*, 2011; Wang *et al.*, 2010).

Compression-induced injury is frequently encountered during handling, packaging, transporting, and storage (Kays, 1991). From grabbing to stacking the bags for display in the market, the RTE leafy vegetables are vulnerable to mechanical damage due to the accidental compression throughout the processes. Spinach is one of the leafy vegetables that have high risk to be mechanical breakage due to its soft and tender properties. It is necessary to examine the changes of mechanical behaviour of leafy vegetables by applying load in order to understand how the mechanical stress damages the plant tissues (Kays, 1991). The mechanical properties of fruits and vegetables can be identified by using compression testing (Bahnasawy *et al.*, 2004; Emadi *et al.*, 2005; Grotte *et al.*, 2001; Sadrnia *et al.*, 2008). Therefore, in this work, compression was selected as an exemplary impact caused during processing and how different spinach varieties respond to it before and after storage was investigated. Microstructural properties of the leaves were also discussed.

MATERIALS AND EXPERIMENTAL

Materials

Spinach (*Spinacia oleracea*) varieties that were used throughout the project were Teen, Organic, and Salad spinach supplied by UK wholesaler and Baby spinach purchased from a local retailer in Birmingham, UK. During transportation, spinach leaves were stored in cool bags with aid of cool packs (6 ± 1°C). Sorting of the leaves was conducted manually to include only good spinach leaves for testing. Good spinach leaves referred to leaves that have no obvious breakage or damage, no signs of disease and no discolouration (yellowing or browning). Samples were stored in a 5 ± 1°C walk-in fridge.

Leaves deformation by uniaxial compression

Compression tests were performed using a mechanical tester, Universal Testing Machine Z030 (Zwick/Roell, Germany). Teen, Organic, Salad and Baby spinach were placed inside an acrylic chamber (110 mm × 150 mm) and undergone uniaxial compression by upper platon. For every condition tested, there were three to five replications of compressions. At maximum the upper platon can compress the samples inside the chamber as the pre-set compression force was noted as minimum tool separation, which was the minimum distance between the upper platon and the bottom of the chamber after the compression (Figure 1).

The objectives are to study the energy of single and multiple loading/unloading compression of bulk spinach, and the energy of multiple loading/unloading compression of stacked spinach. For the studies on single and multiple loading/unloading compression of bulk spinach, Teen, Organic, and Salad spinach were used and compressed under 100 N. The samples were stored for six days. For the study on multiple loading/unloading compression of stacked spinach, only Baby spinach was used and compressed under 50 N, 100 N, and 200 N.

Microscopy imaging technique

The microstructure of the leaves was observed under a light microscope, DM RBE (Leica, Germany). The study focused on analysing the microstructure of different types of spinach samples which included Teen, Organic, and Salad spinach. There were three replicates for each test. About 2 cm of the leaves were finely cut using a sharp disposable scalpel and placed on a glass slide

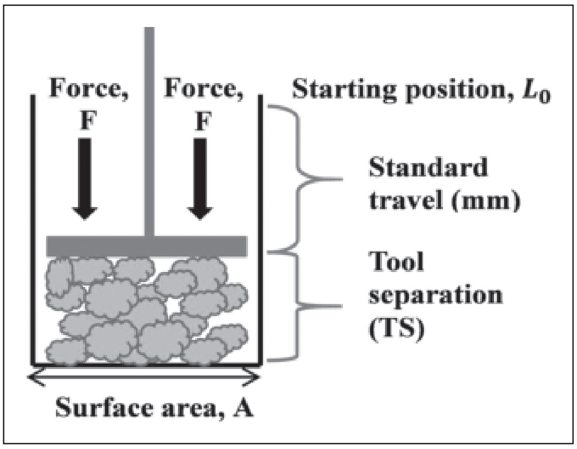

Fig. 1. Illustration of the bulk spinach inside the bed chamber under compression test using Universal Testing Machine (Zwick/Roell UK).

Table 1. MaxW values after compressions before and after storage. The data represent the means of 5 replicates ± standard deviation

	Day 0			Day 6		
	Teen	Organic	Salad	Teen	Organic	Salad
MaxW (J)	2.4 ± 0.3	3.0 ± 0.7	2.1 ± 0.3	1.7 ± 0.2	2.0 ± 0.3	1.2 ± 0.2
AUC (J)	7.8 ± 2.7	8.0 ± 0.4	7.0 ± 1.2	7.0 ± 3.0	7.4 ± 3.5	6.0 ± 0.2

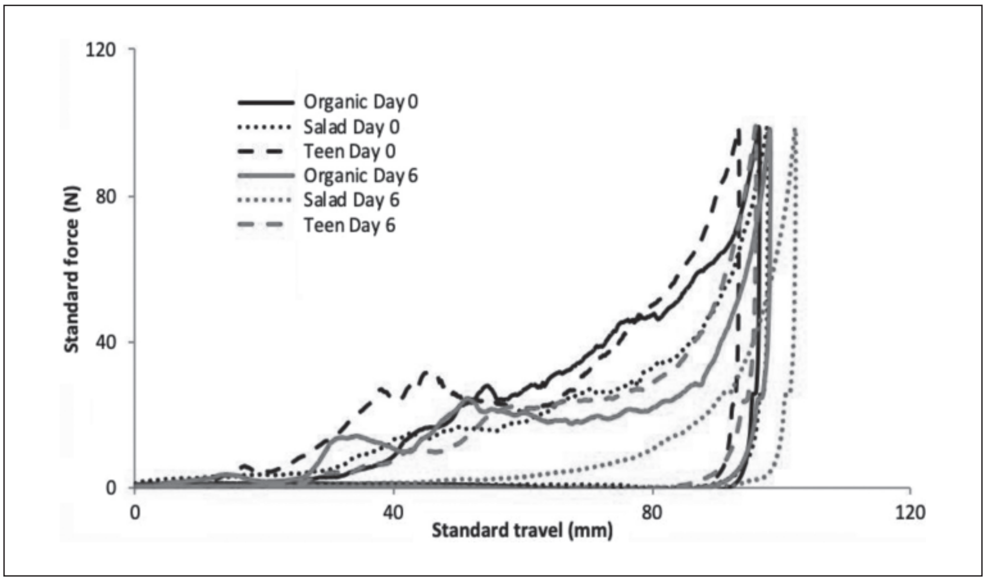

Fig. 2. The force-displacement curves for Organic, Teen and Salad spinach before and after storage.

protected with a thin cover glass. Images were viewed under 10 × 1.0 × 10 objective lens magnification. Cell structure and size of the spinach leaves were analysed and measured using image analysis software, Leica QWin.

RESULTS AND DISCUSSION

The energy of single loading/unloading compression

The maximum work (MaxW) is defined as the maximum work generated from the loading/unloading cycle during the compression. It can also be interpreted as the maximum energy required to compress the leaves. In other words, it can be depicted as the textural behaviour of the leaves. Among the three types of spinach leaves (Organic, Teen, and Salad), the MaxW required to compress Organic spinach was found to be the highest followed by Teen and Salad spinach (Table 1). Besides the MaxW, the area under the curve (AUC) was also calculated. AUC represents the total work required for one complete loading/unloading compression. Again, Organic spinach was shown to have the highest AUC compared to Teen and Salad spinach (Table 1). The MaxW and AUC values tabulated in Table 1 were the average values of five replications of compressions.

The MaxW was found to be decreasing after storage. After storage, the leaves became softer and tender due to the bruising and decay thus less work was required to compress the leaves. Undesirable loads can be the main reason for bruising (Emadi *et al.*, 2009). Energy absorbed during compression is strongly correlated to the bruise volume and thus is a useful parameter to be evaluated in the handling and packing system of fruits and vegetables (Holt & Schoorl 1977; 1982). The AUC also decreased after storage which showed that the total work generated to compress the leaves was reduced due to texture degradation of the product after storage. This can also be seen from the force-displacement curves shown in Figure 2.

Looking from the physical appearance, Organic spinach has round and short shape; Teen spinach has a long-shaped leaf, whereas Salad spinach has sharp-to-the-end-of-tip shape (Figure 3). From the observation in the laboratory, Organic spinach was found to have the roughest and thickest surface,

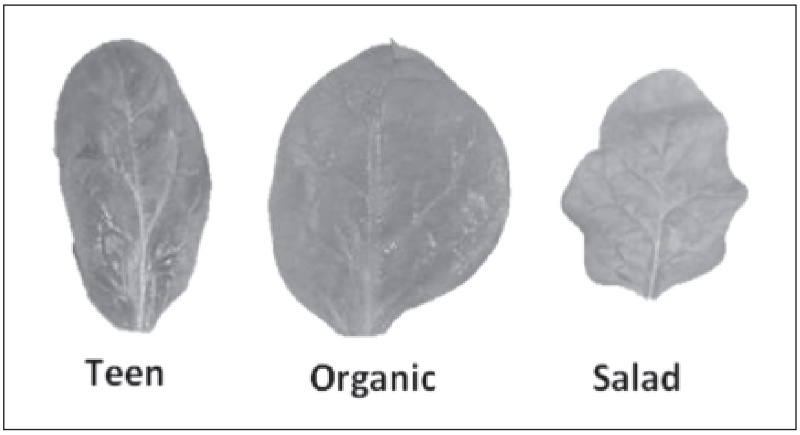

Fig. 3. Images of Teen, Organic and Salad spinach captured using digital camera.

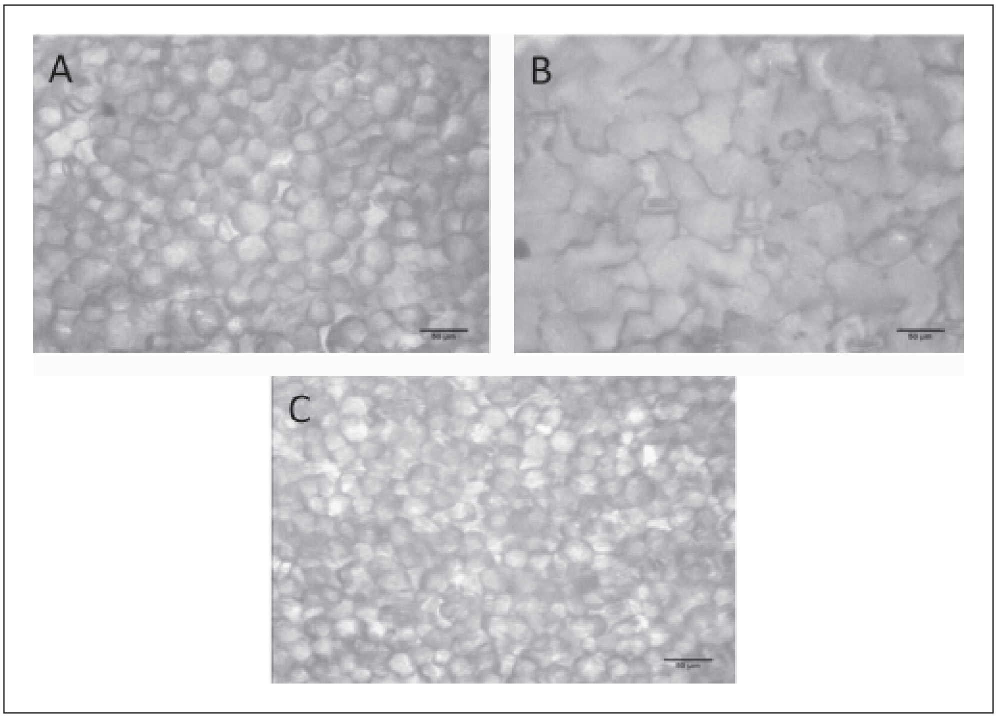

Fig. 4. The microstructures of 3 different types of spinach leaves observed under the light microscope, DM RBE (Leica, Germany); A) Teen, B) Organic, and C) Salad. The average cell sizes were 30.99 µm, 60.75 µm, and 30.10 µm for Teen, Organic, and Salad spinach respectively, was 50 µm.

Teen spinach had medium soft texture and medium-thick leaf whereas Salad spinach had the softest texture and thinnest leaf. Types of cultivar, surface characteristics, stage of maturity, produce size and weight are main factors affecting the mechanical strength of the produce (Kays, 1991).

Apart from that, the intercellular bonding and space are also important components of the tissue strength properties (Shiu *et al.*, 2015). The large intercellular space in spinach leaf provides room for the cells to reorient during compression thus can change the tissue volume significantly (Kays, 1991). The intercellular structures of the spinach leaves showed that Organic spinach has irregular and larger cell sizes, Teen spinach and Salad spinach have round-shaped cells where Teen spinach has larger cell size compared to Salad spinach (Figure 4). The irregular shape of the Organic spinach cells may contribute to the ability of the tissue to resist mechanical stress exerted during the compression. A molecule's shape plays an important role in determining the physical and mechanical properties of the tissues and the way they interact (Silberberg, 2000; Tro, 2008).

The energy of multiple loading/unloading compressions

Apart from single loading/unloading compression, the bulk spinach was also compressed under multiple consecutive compressions. Previous study on the effect of degree of compression towards texture profile of apple, carrot, frankfurter, pretzels, and cream cheese have been conducted (Bourne & Comstock, 1981). The authors controlled the extension cycle to certain range of percentages correspond to the distance between the plates in order to give different degree of compression towards the sample (Bourne & Comstock, 1981). However, there was so much variability for each commodity that the method may not be applicable to other compression speeds. It is necessary to standardise the degree of compression and to report the degree of compression used. Having a standard method could be applicable with other commodities. Plus, previous studies on the effect of degree of compressions towards mechanical properties of fruits and vegetables only focused on the individual samples (Emadi *et al*., 2009; Shiu *et al*., 2015; Thybo & Nielsen, 2000; Zhu & Melrose, 2003). Study on the effect of degree of compression towards mechanical properties of bulk sample as what RTE leafy vegetables were usually displayed in the market is still lacking in the literature.

In this study, the bulk spinach was compressed five times consecutively to create the standard degree of compression (Figure 5). The standard degree of compression referred to the constant starting position before the compression as well as the height of the bulk sample. The speed of each compression was kept constant at 0.5 mm/s. The higher the force, the longer the time required for one complete cyclic loading/unloading compression (i.e. the higher the force, the farther the distance travelled by the piston to compress the sample). For example, with constant sample weight, the time required for one complete compression was 135.9 s, 197.6 s, and 214.0 s for sample compressed under 50 N, 100 N, and 200 N respectively.

Four replications were conducted for each variable introduced in this experiment. As the number of compressions increased, the MaxW decreased (Figure 6 and Table 2). Similar trend was observed at day 6. Another thing to look at is the MaxW for all the three spinach types were the same at the 5th compression both before and after storage. This shows that regardless of the spinach types, which, one type might be rougher or tender than the others, after been compressed consecutively, they generated similar MaxW which showed that they have reached the maximum resistance towards stress. This shows that leaf fracture depends on the degree of compression where higher degree of compression gave more prominent effect (Bourne & Comstock, 1981). As the leaves were compressed further, the cell wall that is near to the contact surface reach the elasticity limit (Thybo & Nielsen, 2000). The energy is dissipated through the cell breakage or stored by distension of the elastic membrane (Holt & Schoorl, 1977). Again,

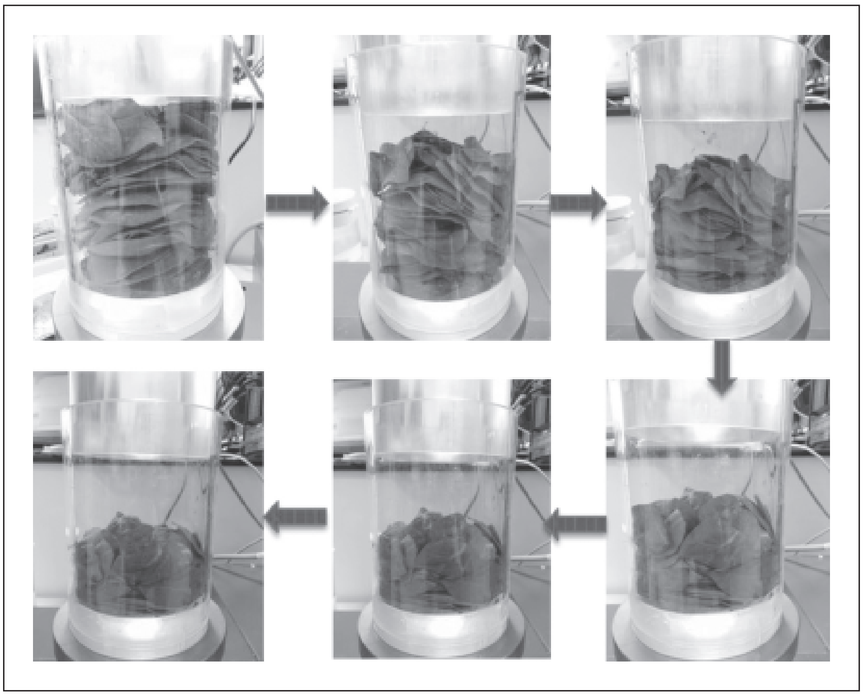

Fig. 5. Randomly thrown bulk spinach before and after 5 times consecutive compressions of 200 N.

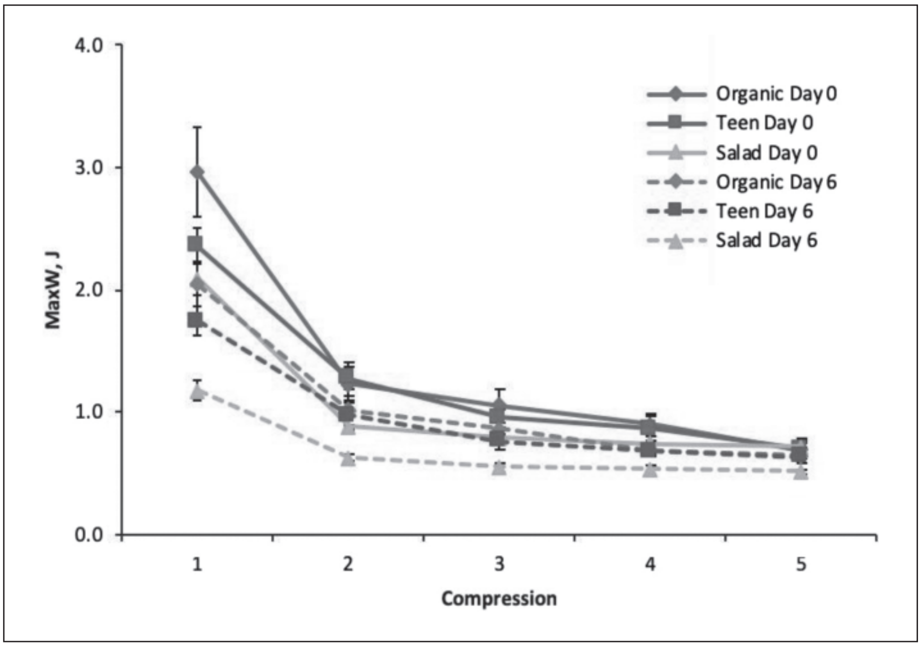

Fig. 6. MaxW of five consecutive compressions before and after storage for Organic, Teen, and Salad spinach.

Table 2. Average MaxW of each five consecutive compressions. The data represent the means of 4 replicates ± standard deviation

Compression	MaxW (J)					
	Day 0			Day 6		
	Teen	Organic	Salad	Teen	Organic	Salad
1st	2.4 ± 0.3	3.0 ± 0.7	2.1 ± 0.3	1.7 ± 0.2	2.0 ± 0.3	1.2 ± 0.2
2nd	1.3 ± 0.3	1.2 ± 0.3	0.9 ± 0.0	1.0 ± 0.2	1.0 ± 0.4	0.6 ± 0.1
3rd	1.0 ± 0.2	1.1 ± 0.3	0.8 ± 0.1	0.8 ± 0.1	0.9 ± 0.3	0.5 ± 0.1
4th	0.9 ± 0.2	0.9 ± 0.2	0.7 ± 0.1	0.7 ± 0.1	0.7 ± 0.1	0.5 ± 0.1
5th	0.7 ± 0.2	0.7 ± 0.1	0.7 ± 0.1	0.6 ± 0.0	0.6 ± 0.1	0.5 ± 0.1

the energy required to compress all types of the spinach leaves after storage was found to be decreasing due to the texture degradation of the leaves after storage.

The energy of multiple loading/unloading compressions of stacked spinach

The work on the multiple compressions was further conducted by stacking the spinach leaves instead of throwing the bulk spinach into the chamber. The purpose was to study the behaviour of the sample at different arrangement and to investigate the resistance between different parts of the sample towards stress. Here, only Baby spinach leaves were used where they were categorised into leaves with and without stem. Twenty leaves were stacked and carefully placed inside the compression chamber. Three replications were conducted for each variable introduced in this experiment.

Results show that, under 200 N compression, leaves with stem required higher energy to compress compared to leaves without stem (Figure 7 and Table 3). However, for leaves compressed under 50 N and 100 N, the difference was only noticed on the first compression and showed similar MaxW between leaves with stem and without stem starting from the 2^{nd} compression till the 5^{th} compression. The resistance towards mechanical damage varies between different component parts of a plant (Kays, 1991). As the compression force increased, the MaxW increased as well (Bourne & Comstock, 1981). The MaxW values reported in this study were lower than the MaxW values reported on the study of multiple compressions on bulk spinach. This is because, the spinach used in this particular objective was Baby spinach which was harvested at early stage where the leaves were smaller and more tender (Jensen, 2017).

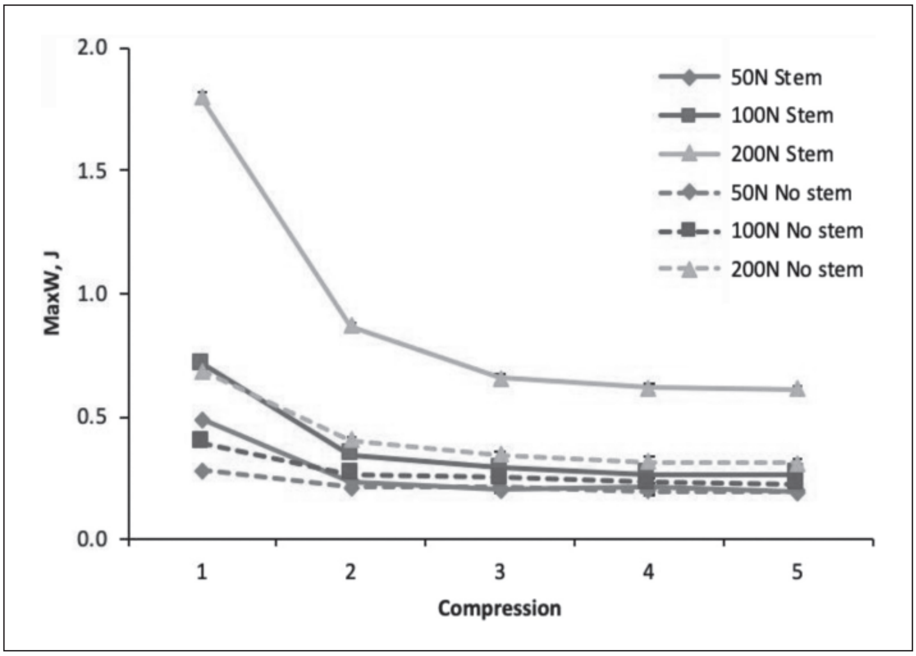

Fig. 7. MaxW of five consecutive compressions before and after storage for stacked Baby spinach with and without stem.

Table 3. Average MaxW of stacked Baby spinach. The data represent the means of 3 replicates ± standard deviation

Compression	MaxW (J)					
	Stem			Without stem		
	50N	100N	200N	50N	100N	200N
1st	0.5 ± 0.0	0.7 ± 0.01	1.8 ± 0.03	0.3 ± 0.01	0.4 ± 0.01	0.7 ± 0.03
2nd	0.2 ± 0.01	0.3 ± 0.02	0.9 ± 0.02	0.2 ± 0.01	0.3 ± 0.02	0.4 ± 0.02
3rd	0.2 ± 0.01	0.3 ± 0.03	0.7 ± 0.03	0.2 ± 0.02	0.2 ± 0.03	0.3 ± 0.03
4th	0.2 ± 0.03	0.3 ± 0.03	0.6 ± 0.04	0.2 ± 0.03	0.2 ± 0.03	0.3 ± 0.03
5th	0.2 ± 0.03	0.3 ± 0.01	0.6 ± 0.03	0.2 ± 0.02	0.2 ± 0.01	0.3 ± 0.03

CONCLUSION

Different spinach varieties respond differently when exposed to stress. From single loading/unloading compression, the energy of compression varies between different types of spinach. The maximum work, MaxW required to compress the spinach was found to be the highest for Organic spinach. This shows that Organic spinach was found to be the best in resisting stress and damage compared to Teen, and Salad spinach. The MaxW was found to be decreasing after storage. This is due to the leaves that became softer after storage thus the energy required to compress the leaves decreased. Also, as the storage day increased, the area under the curve, AUC which represents the springiness of the bulk sample decreased as well. This is due to the bulk sample that became wet after storage and got more attached to each other inside the bag. From the multiple loading/unloading compressions, regardless of spinach variety, they reached maximum resistance towards stress after the 5th compression. From the energy of stacked spinach, it can be suggested that leaf with attached stem resist the stress better than leaf without stem. Suggestion could be made to the fresh vegetable industries during the selection process, to help them choose leaf criteria, that is best in resisting stress.

This work provides basis to understand the effect of compression towards mechanical and microstructural properties of spinach, which can be used to improve the processing and handling of the spinach at the distribution chain and retailer level.

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