Characteristics of Edible Film Based on Okra Slime with the Addition of Amylum Maydis and Glycerol

Warkoyo Warkoyo[1], *Devi* Dwi Siskawardani[1,*], *Sukardi* Sukardi[1], *Damat* Damat[1], *Ivar* Zekker[2], and *Ratri* Juwita Safitri[1]

[1]Department of Food Technology, University of Muhammadiyah Malang, Jl. Raya Tlogomas No. 246, Malang 65144, East Java, Indonesia
[2]Institute of Chemistry, University of Tartu, Ravila 14a 50411 Tartu, Estonia

Abstract. Edible film from polysaccharides has the advantages of aesthetic appearance, oxygen barrier and reduced respiration rate in fruits and vegetables. Okra slime has potential as a stabilizing agent, thickener, and binding agent. While amylum maydis from corn starch contains 27 % amylose and its suitable for edible film, however, it is brittle and rigid. For this reason, formulations with polyol compounds such as glycerol with hydrophilic and hygroscopic characteristics are very useful. This study aims to determine the interaction between amylum maydis and glycerol concentration on the physical characteristics of edible okra slime film. Randomized block design of factorial with two factors and three replication was applied. The first factor was the concentration of amylum maydis [2 %; 2.7 %; 3.3 % (w v^{-1})]. The second factor was glycerol concentrations [1 %; 1.3 %; 1.7 % (w v^{-1})]. The results showed that an interaction between amylum maydis and glycerol concentration on solubility and transparency. The concentration of amylum maydis separately significantly affected thickness, solubility, transparency, tensile strength, and elongation. While the concentration of glycerol separately had a significant effect on solubility and transparency tests. The best treatment was P2F3 (amylum maydis 2.7 % and glycerol 1.7 %).

Keywords: *Abelmoschus esculentus* L., corn starch, eco-friendly packaging, environmentally-friendly.

1 Introduction

Okra slime [*Abelmoschus esculentus* (L.) Moench] has high potential as a basic ingredient for producing edible films. It was proven to have characters that meet the standards. Okra slime had potential as a stabilizing agent, thickener, and binding agent [1, 2]. But it had weakness of physical characteristics, therefore the addition of corn (*Zea mays* L.) starch as supporting ingredient due to its hygroscopic properties at a Relative Humidity (RH) which

* Corresponding author: devi@umm.ac.id

is about 11 %, compared to 13 % cassava starch, 14 % rice starch, and 18 % potato starch. In addition, corn starch contains 27 % amylose that plays a role in the flexibility and strength of the edible film. Furthemore, amylum maydis that was produced from corn starch contains zein which had the ability to form a rigid, glossy, adhesive and grease resistant film [3].

Moreover, edible films made from polysaccharides (carbohydrates) had several advantages and disadvantages. The advantages, namely an aesthetic appearance, ability to act as a barrier to oxygen, can play a selective role in the exchange of O_2 and CO_2 gases resulting of reducing respiration rates in fruits and vegetables. Nevertheless, the disadvantages of edible films from starch, were easily torn or damaged due to low resistance of water and low barrier properties of water due to hydrophilic. In addition, the mechanical properties of starch films were also deficient, such as low elasticity. To improve its characteristics, starch should be mixed with a biopolymer which acts as a plasticizer [4, 5]. Glycerol is a polyanhydrous alcohol compound with three hydroxyl groups. Glycerol was effectively used as a plasticizer on hydrophilic films because it produced a more flexible and smoother edible film [6, 7].

This research purpose was to identify the effect of amylum maydis and glycerol addition on the characteristics of edible film as a substitute for plastic packaging. Eco-friendly packaging is needed because plastic waste is a major global environmental problem [8–11].

2 Methods

Randomized block design factorial with two factors and three replications were used. First factor (P) was the concentration of amylum maydis and second factor (F) was the concentration of glycerol. The combination of independents variable resulted nine treatments, each combination was repeated three times. Analysis of Variance (ANOVA) was made to obtain conclusions of the treatments effected on each parameter. Duncan multiple range test was applied for further test.

The research was consisted of four stages. The first step was okra slime extraction, the second step was corn starch production, then edible film formulation and production, and the last step was parameters analysis. Extraction of okra slime was started with sorting by selecting good fruit which it was not damaged or rotten, then washed thoroughly. The clean okra was cut into 1 cm, then soaked for 8 h to 10 h at cold temperature with a ratio of okra and water 1:6 (w v^{-1}) in 1 000 mL of distilled water. The filtering was used to separate the gel from waste [12].

The corn starch production was started with sorting, peeled, and washed. The corn was separated from the cob, then soaked using distilled water for 48 h and followed the corn kernels were drained and milled. The milling results were deposited for 24 h to obtain corn starch, then filtered to separate the sediment from distilled water. The corn starch precipitated was dried using a cabinet dryer at a temperature of 50 °C for 24 h [13].

Production of edible films were started with homogenization of 150 mL okra slime with corn starch [2 %, 2.7 %, 3.3 % (w v^{-1})], and glycerol [1 %, 1.3 %, 1.7 % (v v^{-1})]. Followed with heating at temperature of 75 °C to 80 °C for 15 min, then cooling to 45 °C, and pouring on a 22 cm × 15 cm × 3 cm pan. The drying steps were in a cabinet dryer with a temperature of 70 °C for 24 h, and cooling at room temperature for 15 min [14].

The resulting edible film were analyzed for thickness; transparency; solubility; tensile strength; elongation; water vapor transmission rate [15, 16].

3 Result and discussion

3.1 Edible film solubility

Based on the analysis of variance, it was a significantly interaction between the concentration of amylum maydis and glycerol on the solubility of the edible film (Table 1). The higher proportion of amylum maydis resulted higher solubility of edible film. This is due to the complex bonding of corn starch-glycerol to form an edible film matrix which is dominated by hydrophilic properties, resulting in an increasing of solubility. Corn starch contains 27 % amylose while potato starch is 22 % and cassava starch is only 17 %. Amylose plays a role in the flexibility and strength of edible films. In addition, corn starch also contains zein which has the ability to inrm a rigid, shiny, abrasion-resistant and grease-resistant film [17–19]. Dissolved solids derived from the basic ingredients for edible films production and the increasing number of molecules in solution caused the highest solubility to indicate that edible films were easily decomposed, applied to instant products, and lower water resistance [19–22].

Table 1. The solubility of edible film.

Treatments	Solubility (%)
Amylum maydis 2 %; Glycerol 1 %	13.19a
Amylum maydis 2 %; Glycerol 1.3 %	13.28a
Amylum maydis 2 %; Glycerol 1.7 %	15.60a
Amylum maydis 2.7 %; Glycerol 1 %	16.48a
Amylum maydis 2.7 %; Glycerol 1.3 %	18.26a
Amylum maydis 2.7 %; Glycerol 1.7 %	18.57a
Amylum maydis 3.3 %; Glycerol 1 %	23.11a
Amylum maydis 3.3 %; Glycerol 1.3 %	33.46a
Amylum maydis 3.3 %; Glycerol 1.7 %	135.16b

Note: The numbers followed with different alphabet showed significantly based on DMRT $\alpha = 5$ %.

3.2 Edible film transparency

According to the analysis of variance, there was a significant interaction between the concentration of amylum maydis and glycerol on the transparency of the edible film. Based on Table 2, transparency of edible film ranges from 4.38 A546 mm^{-1} to 7.42 A546 mm^{-1}. The transparency tends to increase as the concentration of added plasticizer. The higher glycerol in the film resulted in higher transparency. The glycerol ≥ 15 % resulted the bond mobility increased, and when the glycerol ≥ 30 % produced humidity, the tissue expands, and decreased in intermolecular forces and the starch matrix absorbed a lot of water, as the result, the film loses its transparency [23, 17].

The greater concentration of glycerol added, the value of transparency (clarity) tends to increase, which caused the thickness of edible film was increase. It was due to the increasing total amount of dissolved solids in the film. The transparency value decreased, resulted the clarity of the edible film was increased [24, 16]. In addition, the increasing concentration of amylum maydis caused the gelatinization which also affected the transparency of edible film. Transparency was also influenced by the characteristics of raw materials *i.e.,* corn starch had a bright white color. Therefore, starch that had a high brightness would form a more transparent edible film.

Table 2. The transparency of edible film.

Treatments	Transparency (A_{546} mm^{-1})
Amylum maydis 2 %; Glycerol 1 %	4.38a
Amylum maydis 2 %; Glycerol 1.3 %	4.54a
Amylum maydis 2 %; Glycerol 1.7 %	4.94ab
Amylum maydis 2.7 %; Glycerol 1 %	5.08abc
Amylum maydis 2.7 %; Glycerol 1.3 %	5.18abc
Amylum maydis 2.7 %; Glycerol 1.7 %	6.18bcd
Amylum maydis 3.3 %; Glycerol 1 %	6.34bcd
Amylum maydis 3.3 %; Glycerol 1.3 %	6.56cd
Amylum maydis 3.3 %; Glycerol 1.7 %	7.42d

Note: The numbers followed with different alphabet showed significantly based on DMRT $\alpha = 5$ %.

3.3 Edible film thickness

Based on the analysis of variance, there was no interaction between the addition of amylum maydis and glycerol on the thickness. Separately, the addition of amylum maydis concentration had a very significant effect on the thickness of the edible film (Table 3). The composite edible film produced in this study has met the standards of the Japan International Standard 1997 which is around 0.25 mm [25, 26].

The thickness of edible film increased with the concentration of amylum maydis which caused increasing the polymer making up the film matrix, and greater the total solids and resulted thicker edible film. If the concentration of the components of edible film was higher, the total solids in the edible film after drying also increase, therefore could produce a thicker film matrix. The concentration of amylum maydis showed that all the hydrophilic groups in glycerol and corn starch were perfectly bonded. Amylum maydis, and glycerol were a hydrophilic or polar phase. The complex bonds produced by corn starch affect the total solids of the composite edible film matrix.

Table 3. The thickness of edible film.

Treatments	Thicknes (mm)
Amylum maydis 2 %	0.21a
Amylum maydis 2.7 %	0.23ab
Amylum maydis 3.3 %	0.26b
Glycerol 1 %	0.22
Glycerol 1.3 %	0.23
Glycerol 1.7 %	0.25

Note: The numbers followed with different alphabet showed significantly based on DMRT $\alpha = 5$ %.

The thickness of edible film was not significantly increase with the glycerol concentration added. This caused by the difference in glycerol concentration which was not so large, and glycerol had hydrophilic properties that could absorb water vapor around it. In addition, glycerol had the property of binding to starch which forms a starch-glycerol polymer, so the bond between starch and starch was replaced with starch-glycerol in a film matrix.

3.4 Edible film tensile strength

The analysis of variance showed that there was no interaction between the addition of amylum maydis and glycerol on the tensile strength. Separately, the addition of amylum maydis concentration had a very significant effect on the tensile strength of the edible film (Table 4).

The greater concentration of amylum maydis, resulted more hydrogen bonds contained in the film matrix and the chemical bonds of the film would be stronger and difficult to break, because it requires a large amount of energy to break these bonds. The bioplastic particles lead to many physical changes due to more homogeneous and have a tight structure, with these characteristics, produced greater tensile strength and the percentage of elongation. Japanesse Industrial Standard has set a minimum standard for edible film tensile strength was 0.39 Mpa [25, 26].

Table 4. The tensile strength of edible film.

Treatments	Tensile Strength (MPa)
Amylum maydis 2 %	0.64a
Amylum maydis 2.7 %	1.30ab
Amylum maydis 3.3 %	2.04b
Glycerol 1 %	1.83
Glycerol 1.3 %	1.19
Glycerol 1.7 %	0.97

Note: The numbers followed with different alphabet showed significantly based on DMRT $\alpha = 5$ %.

The tensile strength of edible film was not significantly decreased with the glycerol concentration added. This is due to the glycerol molecule would reduce hydrogen bonds and increasing the flexibility of the edible film. If the edible film is made without the addition of glycerol, it will cause the edible film to become brittle. Glycerol could interact with hydrocolloids which will form bond between hydrocolloid and glycerol, result in increased elasticity of an edible film [27–31, 3].

3.5 Edible film elongation

The analysis of variance showed that there was no interaction between the addition of amylum maydis and glycerol on the elongation of edible film. Separately, the addition of amylum maydis concentration had a very significant effect on the elongation of the edible film (Table 5).

Table 5. The elongation of edible film.

Treatments	Elongation (%)
Amylum maydis 2 %	59.5a
Amylum maydis 2.7 %	65.4a
Amylum maydis 3.3 %	79.5b
Glycerol 1 %	71.92
Glycerol 1.3 %	71.97
Glycerol 1.7 %	72.06

Note: The numbers followed with different alphabet showed significantly based on DMRT $\alpha = 5$ %.

The highest percentage of edible film elongation was in the addition of 3 % amylum maydis concentration. This was due to the complex bond in the composite edible film matrix with higher corn starch concentration. The decrease in elasticity was caused by the decreasing distance between the intermolecular bonds, the saturation point had been exceeded and the excess plasticizing molecules were in a separate the polymer phase and will reduce the intermolecular forces among chains, causing to move more freely and flexibility increases (more elastic).

The addition of glycerol concentration did not have a significant effect on the elongation of the edible film. In general, the addition of glycerol as a plasticizer, the molecules in the solution are located between the biopolymer bond chains and can interact by forming hydrogen bonds in the bond chains between polymers, causing the interaction between biopolymer molecules to decrease. Glycerol can cause swelling of the film, so that the bonds between molecules become tenuous, the movement of the polymer chains becomes freer [3, 27–30].

Strong hydrogen bonds are formed between starch molecules and glycerol then become saturated. Corn oat-based edible film using variations of glycerol as a plasticizer, the glycerol concentration will affect the thickness of the edible film, the more the concentration of glycerol in the edible film solution increases, the thickness value increases due to the number of total solids present in the edible film, when the edible film is heated using an oven. Then the evaporation process will occur, and the total solids will settle to the teflon surface which affects the thickness of the edible film. The addition of glycerol can reduce hydrogen bonds in starch molecules, so that the distance between the biopolymer molecules becomes tenuous. With the gap between the biopolymer molecules will increase the flexibility of the film [27–34, 3].

3.6 Edible film WVTR

The analysis of variance showed that there was no interaction between the addition of amylum maydis and glycerol on the elongation of edible film (Table 6). The addition of a greater concentration of corn starch increased the rate WVTR of edible film. This due to corn starch is a polysaccharide that has hydrophilic properties in the film matrix. The hydrogen bonds formed resulted in an increased of matrix film formed, thereby increasing WVTR of the edible film. Increasing the number of solid granules in a polymer reduced the intercellular space of the gel formed.

The addition of glycerol had no significant effect on the WVTR of edible film. This was influenced by the proportion of glycerol used, it would affect the ability of the edible film that accommodate a greater amount of water than the edible film that used a lower concentration of glycerol. The glycerol was a hydrophilic material that was able to increase hygroscopicity which had the ability to absorb water around the edible film. Glycerol was effective as a plasticizer because it was able to reduce internal hydrogen bonds in the intermolecular bonds to soften the film structure, increase the mobility of the biopolymer chain and improve the mechanical properties of the film [3].

Table 6. The water vapour transmission rate of edible film.

Treatments	WVTR (g m^{-2} d^{-1})
Amylum maydis 2 %	1.84[a]
Amylum maydis 2.7 %	1.85[a]
Amylum maydis 3.3 %	2.32[a]
Glycerol 1 %	2.12[a]
Glycerol 1.3 %	2.02[a]
Glycerol 1.7 %	1.88[a]

Note: The numbers followed with different alphabet showed significantly based on DMRT α = 5 %.

The best treatment refers to the edible film standard, namely JIS (Japanese International Standard) [25, 26]. The JIS standard is based on several parameters, namely, thickness, tensile strength, elongation and WVTR. The best treatment edible film okra slime with the addition of corn starch (amylum maydis.) and glycerol was P2F3 (2.7 % amylum maydis and 1.7 % glycerol). The characteristics of P2F3 were shown in Table 7. The solubility

(23.11 %) which indicated the edible film would be easy to digest and transparency (4.38 A546 mm^{-1}) defined that edible film was transparent.

Table 7. The comparison of the best treatment and the Japanese international standard.

Parameter	The best treatment	JIS
Thickness	0.23 mm	Maks 0.25 mm
Tensile strength	0.26 Mpa	Min 0.39 MPa
Elongation	79.5 %	< 10 % very bad; < 50 % very good
WVTR	2.82 g m^{-2} d^{-1}	7 g m^{-2} d^{-1}
Solubility	23.11 %	-
Transparency	4.38 A546 mm^{-1}	-

4 Conclusion

There was an interaction between the concentration of amylum maydis and glycerol on the solubility and transparency of edible film. Separately, the addition of amylum maydis significantly affected the thickness, solubility, transparency, tensile strength, and elongation of the edible film. The best treatment was obtained to P2F3 (amylum maydis 2.7 % and glycerol 1.7 %) with a thickness 0.23 mm, tensile strength 0.26 MPa, elongation 79.5 %, WVTR 2.82 g m^{-2} d^{-1}, solubility 23.11 %, and transparency 4.38 A546 mm^{-1}.

References

1. A. Roy, S.L. Shivastava, S.M. Mandal, Plant Sci. Today, **1**,3: 121–130 (2014) http://dx.doi.org/10.14719/pst.2014.1.3.63
2. W.B. Akanbe, A.O. Togun, J.A. Adediran, E.A.O. Ilupeju, Am. Eurasian J. Sustain. Agric., **4**,1: 1–13 (2010) http://www.aensiweb.net/AENSIWEB/aejsa/aejsa/2010/1-13.pdf
3. D.D. Siskawardani, W. Warkoyo, A.A.P. Siwi, Food Technol. Halal Sci., **3**,1: 26–33 (2020) https://doi.org/10.22219/fths.v3i1.13057
4. H. Hafnimardiyanti, M.I. Armin, Der Pharmacia Lettre, **8**,19: 301–308 (2016) https://www.scholarsresearchlibrary.com/articles/effect-of-plasticizer-on-physical-and-mechanical-charateristics-of-edible-film-from-mocaf-flour.pdf
5. J.A. Borges, V.P. Romani, W.R. Cortez-Vega, V.G. Martins, Int. Food Res. J., **22**,6: 2346–2351 (2015) http://www.ifrj.upm.edu.my/22%20(06)%202015/(24).pdf
6. M. Dick, T.M.H. Costa, A. Gomaa, M. Subirade, A.de Oliveira Rios, S.H. Flôres, Carbohydr. Polym., **130**: 198–205 (2015) https://doi.org/10.1016/j.carbpol.2015.05.040
7. D.D. Siskawardani, W. Warkoyo, R. Hidayat, Sukardi, IOP Conf. Ser.: Earth Environ. Sci., **458**,012039: 1–7 (2020) https://doi.org/10.1088/1755-1315/458/1/012039
8. W. Warkoyo, D.D. Siskawardani, A.Y.P. Siwi, M. Wachid, I. Zekker, J. Onthong, Sarhad J. Agric., **37**,Special Issue 1: 144–152 (2021) https://dx.doi.org/10.17582/journal.sja/2021.37.s1.144.152
9. I. Iswahyudi, W. Widodo, W. Warkoyo, R.H. Setyobudi, A. Sutanto, Z. Vincēviča-Gaile, et al., E3S Web of Conf., **432**,00015: 1–13 (2023) https://doi.org/10.1051/e3sconf/202343200015
10. I. Iswahyudi, W. Widodo, A. Sutanto, R.H. Setyobudi, R. Tonda, E.D. Purbajanti, et al., Proc. Pak. Acad. Sci.: B (2023) In Press.
11. I. Iswahyudi, W. Widodo, W. Warkoyo, R.H. Setyobudi, D. Damat, D. Roeswitawati, et al., Sarhad J. Agric., (2023) In Press.

12. M. de A.P. Cotrim, A.C. Mottin, E. Ayres, Macromol. Symp., **367**,1: 90–100 (2016) https://doi.org/10.1002/masy.201600019
13. M. Djaeni, S. Sumardiono, S. Suherman. *Advance Drying Technology for Heat Sensitive Products*, Semarang: Undip Press (2014) https://scholar.google.com/scholar?hl=en&as_sdt=0%2C5&q=Advance%20drying%20technology%20for%20heat%20sensitive%20products
14. O. Ismail, A.S. Kipcak, I. Doymaz, Athens J. Sci., **6**,3: 155–168 (2019) https://doi.org/10.30958/ajs.6-3-1
15. ASTM D 368 M-III. *Standart Test Method for Tensile Properties of Thin Plastic Sheeting.* West Conshohocken, PA. (2001)
16. A. Al-Hasan, M.H. Norziah, Food Hydrocoll, **26**,1: 108–117 (2012) https://doi.org/10.1016/j.foodhyd.2011.04.015
17. L. Kumar, D. Ramakanth, K. Akhila, K.K. Gaikwad, Environ. Chem. Lett., **20**: 875–900 (2022) https://doi.org/10.1007/s10311-021-01339-z
18. H. Chen, Z. Sun, H. Yang, Sci. Hortic., **244**: 157–164. (2019) https://doi.org/10.1016/j.scienta.2018.09.039
19. T.J. Gutierrez, K. Alvarez, J. Funct. Food, **26**,2: 750–762 (2016) https://doi.org/10.1016/j.jff.2016.08.054
20. M.D. Hazrol, S.M. Sapuan, E.S. Zainudin, M.Y.M. Zuhri, N.I.A. Wahab, Polymers, **13**,2: 1–22 (2021) https://doi.org/10.3390/polym13020242
21. M.I. Pinzon, O.R. Garcia, C.C. Villa, J. Sci. Food Agric., **98**,11: 4042–4049 (2018) https://doi.org/10.1002/jsfa.8915
22. M.W. Apriliyani, R.D. Andriani, P.P. Rahayu, P. Purwadi, A. Manab, J. Ilmu dan Teknologi Hasil Ternak, **15**,3: 162–171 (2020) https://doi.org/10.21776/ub.jitek.2020.015.03.4
23. P.F. Munoz-Gimena, V. Oliver-Cuenca, L. Peponi, D. Lopez, Polymers, **15**,13: 1–45 (2023) https://doi.org/10.3390/polym15132972
24. S. Bao, Shiying Xu, Z. Wang, J. Sci. Food Agric., **89**,15: 2692–2700 (2009) https://doi.org/10.1002/jsfa.3775
25. Japanesse Industrial Standart 2 1707. *General Rules of Plastic Film Food Pacaging. Japanesse-Standart Assosiation*. Tokyo: JSA (1997) https://standards.gloalspec.com/std/13385455/JIS%20Z%201707
26. M. Rahmawati, M. Arief, W.H. Satyantini, IOP Conf. Ser.: Earth Environ. Sci., **236**,012129: 1–8 (2019) https://doi.org/10.1088/1755-1315/236/1/012129
27. F. Yulistiani, N. Khairunisa, R. Fitriana. J. Phys.: Conf. Ser., **1450**,012001: 1–6 (2020) https://doi.org/10.1088/1742-6596/1450/1/012001
28. E. Talón, K.T. Trifkovic, V.A. Nedovic, B.M. Bugarski, M. Vargas, A. Chiralt, et al., Carbohydr. Polym., **157**: 1153–1161 (2017) https://doi.org/10.1016/j.carbpol.2016.10.080
29. T.K. Mendy, A. Misran, T.M.M. Mahmud, S.I. Ismail, Sci. Hortic., **246**: 769–776 (2019) https://doi.org/10.1016/j.scienta.2018.11.054
30. D.S. Kumar, D.E. Tony, A.P. Kumar, K.A. Kumar, D.B.S. Rao, R. Nadendla, Int. Res. J. Pharm. Appl., **3**,4: 129–132 (2013) https://scienztech.org/index.php/irjpas/article/view/586
31. S.T.A. Haider, S. Ahmad, A.S. Khan, S.M.A Basra, Pak. J. Agric. Sci., **54**,1: 35–44 (2017) https://www.researchgate.net/publication/316561705_Comparison_of_different_fruit_coatings_to_enhance_the_shelflife_of_Kinnow_Mandarin

32. O. Hafeez, A.U. Malik, A.S. Khan, A. Rehman, Q.A. Javaid, Int. J. Agric. Biol., **14**,1: 47–54 (2012) https://www.researchgate.net/publication/256089277_Impact_of_Different_Packaging_Types_and_Low_Temperature_Shipping_Durations_on_Fruit_Quality_and_Marketability_of_Pakistani_Mangoes
33. M.V. Tzoumaki, C.G. Biliaderis, M. Vasilakakis, Food Chem., **117**,1: 55–63 (2009) https://doi.org/10.1016/j.foodchem.2009.03.076
34. K. Monzon-Ortega, M. Salvador-Figueroa, D. Galvez-Lopez, R. Rosas-Quijano, I. Ovando-Medina, A. Vazquez-Ovando, J. Food Sci. Technol., **55**: 4747–4757 (2018) https://doi.org/10.1007/s13197-018-3397-2