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bilalodin 1 <bilalodin@unsoed.ac.id>

Invitation to Review Manuscript for Molekul

3 messages

Jurnal Molekul <j.molekul@gmail.com> Mon, Sep 5, 2022 at 6:59 PM
To: bilalodin@unsoed.ac.id

Dear **Dr. Bilalodin**,

I am writing to you on behalf of "MOLEKUL", A Scientific Journal in Chemistry. The journal is abstracted and indexed in SCOPUS (Q3), CAS, ACI, SINTA, DOAJ. Please visit our website: http://jos.unsoed.ac.id/index.php/jm/index

Currently, we have received a manuscript (attached) with the title of "**Development and Characterization of Edible Films Based on Gelatin/Chitosan Composites Incorporated with Zinc Oxide Nanoparticles for Food Protection"** for possible evaluation. Your name is identified as a potential reviewer regarding your research activity.
Therefore, I would like to invite you to evaluate the submitted manuscript. Your contribution would help us to improve the quality of the journal and the submitted manuscript. **If you agree to review this article, please reply to this email**.

I also included an abstract for your consideration

The increase in cases of global environmental pollution due to plastic waste makes the development of biodegradable active packaging very urgent. Gelatin (G), is one of the potential edible film raw materials. However, its weak water barrier and mechanical properties have limited its wide application. The addition of chitosan nanofiber (CHNF) and zinc oxide nanoparticles (ZnONP) is expected to improve the mechanical and barrier properties and present antioxidant and antimicrobial properties to the G film. Characterization results using FTIR, SEM, and DSC showed good compatibility between the G, CHNF, and ZnONP matrix. Meanwhile, the packaging test results confirmed that gelatin, CHNF, and ZnONP-based composite films have the potential to be used as functional materials in food packaging.

Best regards,

Senny Widyaningsih
Editor
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Universitas Jenderal Soedirman
website: http://jos.unsoed.ac.id/index.php/jm/index
email : j.molekul@gmail.com

2 attachments

6630-99Z_Article Text-24771-1-2-20220818.doc
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87K

bilalodin 1 <bilalodin@unsoed.ac.id>
To: Jurnal Molekul <j.molekul@gmail.com>
Mon, Sep 5, 2022 at 8:16 PM

Dear Editor

Thank you for trusting me to review the submitted manuscript. I am ready to carry out the task. I immediately review the manuscript.

Best regards
Bilalodin
[Quoted text hidden]

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bilalodin 1 <bilalodin@unsoed.ac.id>
To: Jurnal Molekul <j.molekul@gmail.com>
Wed, Sep 7, 2022 at 10:49 PM

Dear Editor

I send the results of the review article with the title " **Development and Characterization of Edible Films Based on Gelatin/Chitosan Composites Incorporated with Zinc Oxide Nanoparticles for Food Protection".** The parts that need to be corrected are listed in the manuscript. After minor revisions by the author, the article can proceed to the publication process.

Best regards
Bilalodin

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2 attachments

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Development and Characterization of Edible Films Based on Gelatin/Chitosan Composites Incorporated with Zinc Oxide Nanoparticles for Food Protection

Pengembangan dan Karakterisasi Film Edibel Berbasis Komposit Gelatin/Kitosan dengan Nanopartikel Seng Oksida untuk Perlindungan Pangan

Comment [L1]: delete

Asrul Bahar[1], Samik Samik[2], Maria Monica Sianita[2], Nita Kusumawati[2*], Ianatul Khafidlah[2], Supari Muslim[3], AR. Sella Auliya[4]

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ABSTRACT. The increase in cases of global environmental pollution due to plastic waste makes the development of biodegradable active packaging very urgent. Gelatin (G), is one of the potential edible film raw materials. However, its weak water barrier and mechanical properties have limited its wide application. The addition of chitosan nanofiber (CHNF) and zinc oxide nanoparticles (ZnONP) is expected to improve the mechanical and barrier properties and present antioxidant and antimicrobial properties to the G film. Characterization results using FTIR, SEM, and DSC showed good compatibility between the G, CHNF, and ZnONP matrix. Meanwhile, the packaging test results confirmed that gelatin, CHNF, and ZnONP-based composite films have the potential to be used as functional materials in food packaging.

Comment [L2]: Please rearrange the abstract in the following order
1. The urgency of the research topic
2. Research objectives
3. Methods
4. Results

Keywords**:** CHNF, Edible, Gelatin, Packaging, ZnONP

INTRODUCTION

According to a report by the Food and Agricultural Organization (FAO), 14% of food products produced globally are damaged before reaching sales (Susmitha, et al., 2021). Short shelf life due to the susceptibility of dairy products to microbial contamination even before consumption is one of the main problems that trigger waste. The delicious taste and content of several intrinsic nutrients make cheese one of everyday life's most consumed food products. However, cheese is susceptible to the growth of pathogenic Listeria monocytogenes and Staphylococcus aureus, both at processing and storage (Lin, et al., 2019). As a solution, packaging that can extend the shelf life of cheese is used. Most food packaging comes from synthetic plastics which do not biodegrade in nature. The intensive production, application, disposal, and stockpiling of this packaging significantly negatively impacts society and the environment (Liu, et al., 2020). Therefore, it is necessary to develop biopolymer packaging that is cheap, biodegradable, renewable, available in nature, and eco-friendly (Lin, et al., 2019).

Gelatin is a potential biopolymer for the production of biodegradable packaging because it has several advantages, such as good film-forming ability, biodegradability, abundance, high oxygen barrier capacity, as well as low gelling and melting point, and also the ability to act as a functional agent carrier (Amjadi, et al., 2019; Bahar, et al., 2020; Kusumawati, et al., 2019; Bahar, et al., 2018; Bahar, et al., 2019). However, the poor water barrier properties and low mechanical resistance have limited the application of gelatin in food packaging (Quero, et al., 2018). Several steps have been modified to overcome this problem, including using several different compounds to obtain better barrier properties and the incorporation of nanoparticles to enhance the mechanical resistance, and antioxidant and antibacterial properties of the biopolymers (Amjadi, et al., 2019).

Comment [L3]: (G)

Like gelatin, chitosan (CH) biopolymer is also potentially involved in the development of food packaging due to its biocompatibility and non-toxicity (Davoodi, et al., 2020). Of all its physical forms, chitosan nanofibers (CHNF) attract more attention because of their large surface area to volume ratio and high mechanical strength when combined with other polymers. The addition of chitosan nanofibers (CHNF) is a potential step to increase the mechanical resistance while reducing the WVP of gelatin packaging.

Comment [L4]: what does this word stand for

Meanwhile, the addition of zinc oxide nanoparticles is expected to increase the antioxidant and antimicrobial activity of the G-CHNF film to create an active packaging that can provide optimal protection against the spread of the virus. Yadav et al. (2021) reported the broad-spectrum antimicrobial activity of zinc oxide nanoparticles (ZnONPs) (Yadav, et al., 2021). These nanoparticles are also more stable, non-toxic, have a high surface-to-volume ratio, surface reactivity, antioxidant activity, and excellent photocatalytic properties (Kamdem, et al., 2019). The use of ZnONP as a packaging material for food, pharmaceuticals, and cosmetic products has been approved by the United States.

Comment [L5]: Add the research objective at the end of the sentence.

EXPERIMENTAL SECTION

Materials

Comment [L6]: Write down the chemicals used in the research

Antibacterial Activity of Gelatin, CHNF, ZnONP

The agar diffusion method evaluated the gelatin, CHNF, and ZnONP antibacterial activity. The diameter of the inhibition zone formed was measured using a caliper.

Comment [L7]: delete this section because the description is already in the film characterization

Composite Films Preparation ~~Making~~

Comment [L8]: Preparation of Composite Fil

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G, G.CHNF, G.ZnONPs, G.CHNF.ZnONPs films were made using the casting knife method with varying thicknesses (0.8-1.2 mm). The 4% (w/v) gelatin solution was stirred using a hot plate magnetic stirrer (45 ºC; 30 minutes). In a different place, the CHNF solution was prepared with a composition of 10% by weight of dry gelatin, dissolved in 2% acetic acid, and stirred (50 °C; 120 minutes), while the ZnONP solution was prepared with a composition of 5% by weight of dry gelatin, dissolved in 2% acetate acid and stirred (100; 360 min). Glycerol plasticizer prepared with a composition of 25% by weight of dry gelatin, dispersed in a 4% gelatin solution supplemented with previously prepared CHNF, ZnONP, and CHNF-ZnONP (1:1) solutions, then stirred (25 °C; 60 min). To ensure that the film solution was free of air bubbles, the films were degassed by ultrasound at 30 ºC for 30 minutes. The film solution was then cast on an acrylic plate with an area of 14 x 14 cm and dried at 30 ºC for 72 hours. The dry films were kept in a desiccator (50% relative humidity; 25℃) for 72 hours before testing (Amjadi, et al., 2019).

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Film Characterization

The structural interactions of the films were evaluated using Perkin Elmer Spectrum Two FTIR spectroscopy (China). The morphology, surface, and cross-section of the films, were evaluated by FEI Inspect-S50 SEM (Canada). In particular, cross-sectional films were fabricated using Mikrotomy Microm, HM315 (Germany). The thermal stability of the film was evaluated by Linseis STA PT 1600 DSC (Germany) at a temperature of 20-300 ℃. The mechanical parameters of the film were tested using autograph, AG-10TE (Japan). Water absorption was evaluated based on the S. Amjadi (2019) method (Amjadi, et al., 2019). The water vapor permeability of the film was analyzed by the ASTM E96-05 standard method (ASTM, 2005). Transparency and light transmittance, as well as the film color parameters (CieLab coordinates), such as brightness (L*), reddish/greenish (a*), and

Comment [L11]: Fabrication?

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yellowish/bluish (b*) were measured with a Shimadzu UV-2401-PC spectrophotometer (Japan). The antibacterial activity of the film was determined using the agar diffusion method.

RESULTS AND DISCUSSION

Comment [L13]: Please in the results and discussion compared with other people's research

Fourier Transform Infra-Red

All nanocomposite films showed similar specific peaks: 1) amide-A band at 3283.99 - 3289.17 cm^{-1} which also indicates a secondary amine group; and 2) amide-B at 2,936.40 - 2937.38 cm^{-1}. Other specific peaks that also appear in all films are amide-I, amide-II, and amide-III at 1629.54 - 1633.88 cm^{-1}; 1538.64 - 1547.98 cm^{-1}; 1238.38 - 1239.40 cm^{-1}, respectively. The amide-I and II bands indicate the formation of cross-links between the peptide chains. Meanwhile, the amide-III band shows the ability of the protein to maintain the triple helix complex (Cai, et al., 2019).

Comment [L14]: Give information at the beginning of the sentence. The purpose of the evaluation using FTIR and state the results in wha picture.

Comment [L15]: Composite film or nanocomposit film.. consistent.

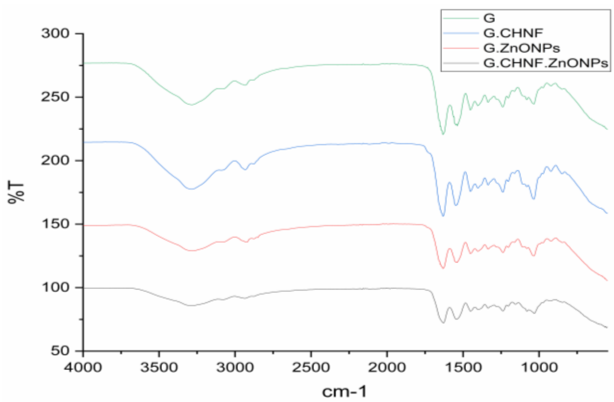

Figure 1. Fourier transform infrared (FTIR) spectrum of ~~film samples with various compositions~~

Comment [L16]: G, G.CHNF, G.ZnONPs, G.CHNF.ZnONP..
please add to the picture the shift direction and pe position of Amide I, II III and domain wavenuml

The addition of CHNF causes a shift in wavenumber, especially in the amide-A, amide-I, amide-II, and amide-III bands due to the formation of intramolecular hydrogen bonds in chitin formed during electrospinning. The addition of ZnONPs also causes a shift due to the formation of hydrogen bonds between the N-H groups on gelatin and ZnONPs. The incorporation of CHNF and ZnONPs also causes a shift in the G.CHNF.ZnONPs film due to the interaction between -OH, -COOH, and $-NH_2$ groups in gelatin with -COO, -OH, and $-NH_2$ groups in CHNF and Zn^{2+} in ZnONPs (Ge, et al. al., 2018). All nanocomposite films also showed interactions with glycerol which were observed at 1033.92 cm^{-1} (G); 1033.29 cm^{-1} (G.CHNF); 1032.28 cm^{-1} (G.ZnONPs); 1032.40 cm^{-1} (G.CHNF.ZnONPs).

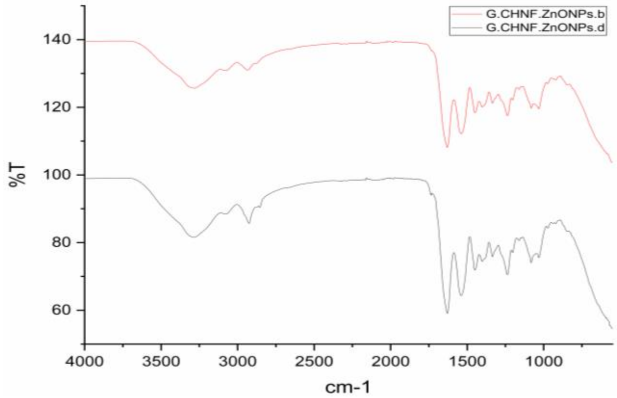

Figure 2. The infrared spectrum of G.CHNF.ZnONPs film with various casting thicknesses: 1.0 mm (b), and 1.4 mm (d)

Comment [L17]: Pictures are given label a an not b and d. label a and b are pasted on the spectr to make it easier to read

Changes in the thickness of the G.CHNF.ZnONPs nanocomposite film triggered a shift in wavenumber, especially in the amide-A band (Figure 2), where at a thickness of 1.0 mm the amide-A band was observed at 3287.26 cm^{-1}, while at a thickness of 1.4 mm the amide-A band was observed at 3286.50 cm^{-1}. This shift is related to the formation of hydrogen bonds between chitin and gelatin molecules (Ge et al., 2018).

Comment [L18]: bold

Scanning Electron Microscopy

The SEM image (Figure 3) shows the surface tends to be rough with cracks and pores in the G film.

Comment [L19]: bold

Comment [L20]: The sentence should start wi The results of Scanning Microscopy (SEM) of G, G.CHNF, G.ZnONPs, and G.CHNF.ZnONP.. are shown in Figure 3. SEM results show that…..

1 paragraph consists of 3 sentences

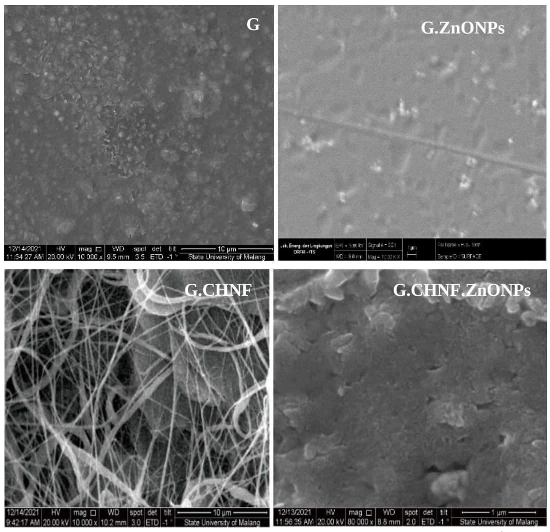

Figure 3. SEM images of ~~the film samples with various compositions~~

Comment [L21]: G, G.CHNF, G.ZnONPs, G.CHNF.ZnONP..

Interconnected pore microstructure changes were detected after the addition of CHNF with filamentous structure. The porous structure facilitates the attachment of ZnONP to the film matrix resulting in increased density and compactness which reduces cracking and porosity and improves mechanical and thermal stability, as well as water barrier capabilities.

Film thickness was also detected to affect the morphology of the G.CHNF.ZnONPs nanocomposite films (Figure 4). An increase in the cast thickness causes an increase in the availability of solids thereby increasing the density and compactness of the film matrix (Ansari, et al., 2018). Figure 4 shows an increase in the surface smoothness of the film as the cast thickness increases.

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Comment [L23]: Start with general sentences. state the results of the SEM then explain

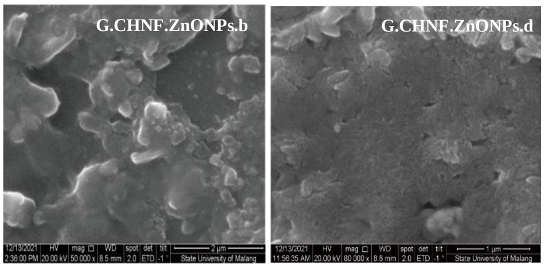

Figure 4. SEM images of the G.CHNF.ZnONPs film with variations in casting thicknesses: 1.0 mm (b), and 1.4 mm (d)

Comment [L24]: replace labels a and b.

Differential Scanning Colorimetry (DSC)

Comment [L25]: *The sentence should start wi The results of* **Differential Scanning Colorimetr**) of G, G.CHNF, G.ZnONPs, and G.CHNF.ZnON are shown in Figure 5. DSC results show that

The thermal analysis showed the appearance of two sharp endothermic peaks from the G film, namely at 124.05 °C which corresponded to the water vapor evaporation and the amino acid-rich region devitrification, and at 222.65 °C which corresponded to the imino-acid-rich region (proline and hydroxyproline) devitrification. The shift of the first (to 139.35 °C) and the second endothermic peak (to 213.45°C) of the G.CHNF film were associated with increased evaporation of moisture content and decomposition of nanofiber, respectively (Ebrahimi, et al., 2019). A further shift was observed in the G.ZnONP film, where the first endothermic peak shifted to 174.55 °C and the second to 224.85 °C. Compared to the other three films, the most distant shift of the two endothermic peaks of the G.CHNF.ZnONP film, i.e. 185.55 for the first peak and 236.15°C for the second showed a synergistic effect of CHNF, ZnONPs, and gelatin increasing the gelatin chain sequence and film crystallinity (Boura-Theodoridou, et al., 2020).

Increasing the casting thickness also tends to increase the thermal resistance of the film. G.CHNF.ZnONPs film with a thickness of 1.4 mm showed a shift of the first

Comment [L26]: (Figure 6

endothermic peak to 185.55 °C and the second to 236.15 °C compared to 1.0 mm. The increase in thickness causes the formation of more crystal structures in the gelatin-CHNF nanocomposite film, one of which is due to CHNF nucleation (Wang et al., 2021).

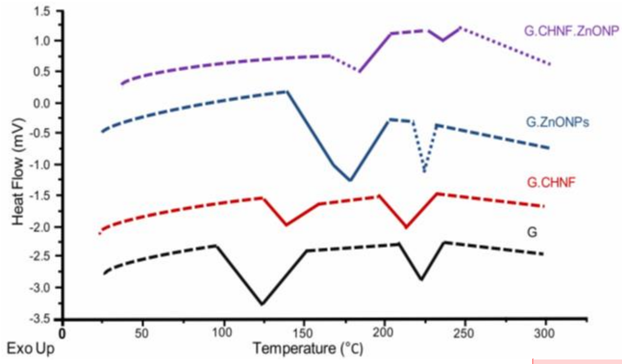

Figure 5. DSC spectrum of ~~film samples with various compositions~~

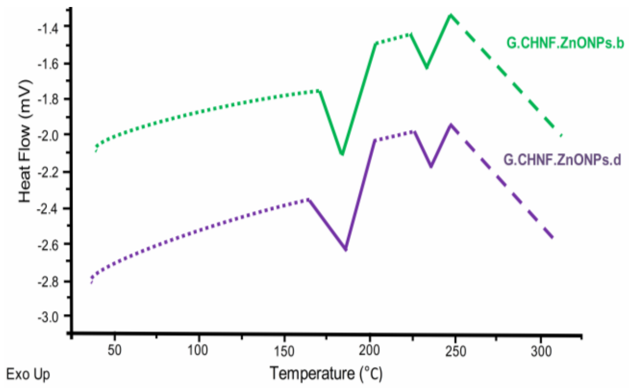

Figure 6. DSC spectrum G.CHNF.ZnONPs film with variations in casting thickness: 1.0 mm (b); and 1.4 mm (d)

Thickness and Mechanical Properties

Figure 7 shows an increase in film thickness after adding ZnONPs and a decrease in film thickness after adding CHNF. The existence of interconnected pore after the addition of CHNF allows the entry of ZnONPs into the polymer matrix resulting in a thinner G.CHNF.ZnONP film than G.ZnONP. Film thickness was also found to increase with increasing casting thickness. The larger the volume of the casting solution, the more total solids in the cast area and the higher the thickness of the resulting film (Hari, et al., 2021).

The mechanical properties of the film were analyzed using the parameters of the tensile strength (UTS), elongation (ETB), and Young's modulus (YM). Base on the Figure 8 shows the higher UTS and YM of G.ZnONPs films than that of G films. ZnONP particle

Comment [L27]: G, G.CHNF, G.ZnONPs, an G.CHNF.ZnONP.
The image is placed under the first paragraph

Comment [L28]: Replace label a and b

Comment [L29]: are showen in Figure 8 dispersion increases the interactions in the matrix and the mechanical properties of the films. On the other hand, the decrease in free volume between polymer chains due to ZnONP dispersion caused the polymer network to be less permeable, resulting in a decrease in ETB from 0.081 to 0.009%. (Nandiwilastio, et al., 2019). In line with this, the increase in crystallinity due to the formation of interconnected pore networks by CHNF created stronger interfacial interactions thereby increasing UTS by 73.21% and YM by 80.33% of G.CHNF films. The presence of hydrogen bonds between chains further strengthens the mechanical stability. The addition of CHNF also caused a decrease in ETB by 0.032% in the G.CHNF film. The increase in intermolecular and intramolecular interactions in the matrix after the addition of CHNF and ZnONP simultaneously resulted in lower flexibility and mobility of polymer segments compared to G.CHNF films (Al.-Tayyar, et al., 2020).

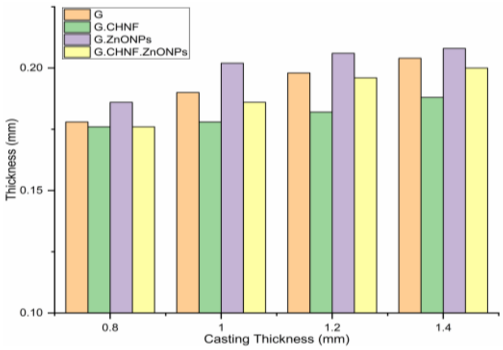

Figure 7. Film thickness with variation of composition and casting thickness

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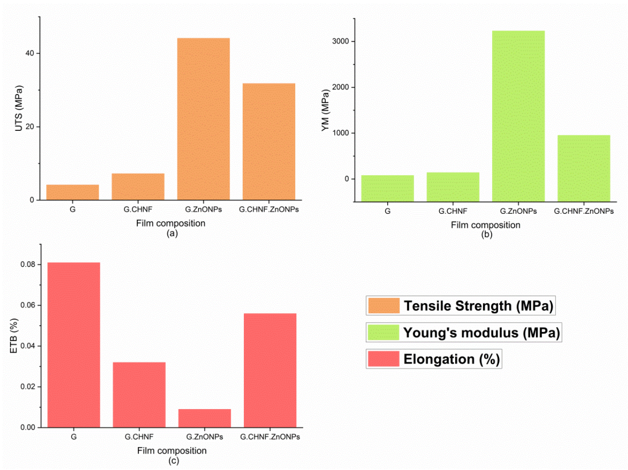

Figure 8. ~~Film~~ mechanical properties of films samples ~~with variation of composition~~

Film thickness also shows an influence on the mechanical parameters of nanocomposite films (Wafi, et al., 2020). The increase in UTS with increasing film thickness is closely related to molecular weight. The closer the distance between the molecules, the stronger the interactions. This condition results in an increase in UTS and a decrease in ETB. ~~Figure 9 shows~~ The effect of thickness on the mechanical parameters of the G.CHNF.ZnONP film is shown in Figure 9.

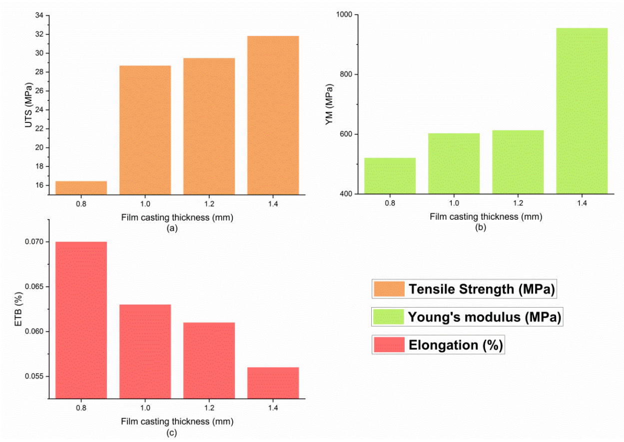

Figure 9. Film mechanical properties with variation of casting thickness

Barrier Properties

Water absorption and water vapor permeability (WVP) are the two main parameters used to study moisture transfer. Figure 10 shows the percentage of water absorption after 96 hours of conditioning at 99% RH. Although there was no significant difference ($p>0.05$) between G.CHNF and G films at the same thickness, G films still showed an increase in the percentage of water absorption with increasing thickness. The addition of ZnONP caused a significant decrease ($p<0.05$) in the percentage of water absorption in the G film and reached its peak after the addition of CHNF and ZnONP. Increased cross-link formation due to the addition of both reduces the mobility of the polymer chain.

The water vapor permeability (WVP) of the film showed the same pattern as the percentage of water absorption. In contrast to the effect of adding CHNF, hydrogen bonds formed between gelatin-ZnONP caused a decrease in free hydrophilicity, thereby reducing the WVP value of G.ZnONP films. The lowest WVP value was produced by G.CHNF.ZnONP film. These results indicate a more significant effect of adding ZnONP than CHNF in decreasing WVP (Soradech, et al., 2017). The decrease in gelatin chain

Comment [L31]: , The moisture absorption an WVP values of the nanocomposit are shown in Fi 10
correct the next sentence to match

mobility resulted in a decrease in WVP properties and an increase in the water barrier film simultaneously.

An increase in thickness was found to increase the hydrophilicity of the film. The increase in thickness produces strain which makes the matrix more brittle and prone to cracking. Under these conditions, water vapor flows faster through the resulting crack.

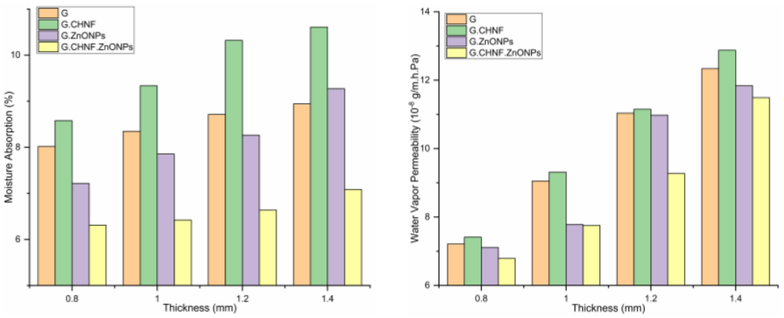

Figure 10. Film's moisture absorption (a); and WVP (b) with variation of composition and casting thickness

Comment [L32]: Image quality needs to be improved so that the x and y axis descriptions are clearer

Transparency

Transparency is an important parameter for verifying the quality of packaging materials and the ability to block UV rays that cause deterioration in food product quality due to spoilage. Figure 11b shows the transparency values for each film. Although not significant ($p>0.05$), G.CHNF film was found to have lower transparency than G film. The CHNF filament structure causes a decrease in light scattering on the CHNF-gelatin surface (Chen, et al., 2018). The dense and complex structure of ZnONP in the matrix further decreases light scattering and increases film opacity (Ngo, et al., 2018). The combination of CHNF and ZnONP showed a synergistic effect which resulted in a maximum decrease in the transparency of the G.CNF.ZnONP film. In general, the G film in this study, either with or without the addition of CHNF and ZnONP or a combination of both, has lower transparency than commercial packaging, such as Low-Density Polyethylene (LDPE) (transparency value 4.26) and oriented polypropylene (transparency value 1.57).

Thickness also shows the same effect on film transparency. Increasing the thickness will increase the number of layers formed thereby reducing light transmission. The more layers of atoms that make up the film will trigger an intensification of the collision with the light particles, thus creating a significant barrier to transmission into the package (Roy & Rhim, 2021).

Comment [L33]: film transmission?

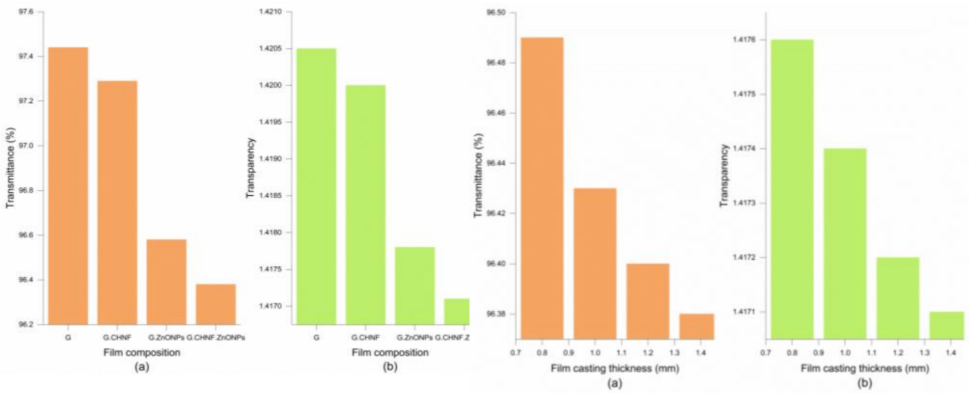

Figure 11. Film's transmittance (a) and transparency (b) with a variation of composition (1) and casting thickness (2)

Comment [L34]: Image quality needs to be improved so that the x and y axis descriptions are clearer

Color Properties

The color properties of the film greatly affect the appearance of product packaging and consumer acceptance of the product. The color properties parameters (L*, a*, b*, E, WI, and YI) of the film are shown in Table 1. High collagen content causes the G film to have a clear color (Yusof & Zain, 2019). A decrease in the L* value of G films was detected after CHNF addition. The CHNF filament structure reduces light scattering on the CHNF-gelatin surface, resulting in a decrease in transparency and L* value (Chen, et al., 2018). A further significant decrease ($p < 0.05$) was detected after ZnONPs addition. The dense and complex structure of ZnONPs inhibits light transmission and causes a decrease in transparency and L* value (Al-Tayyar, et al., 2020).

Comment [L35]: bold

G.CHNF film produced lower a* and higher b* values than G film, 0.26 and -0.34, respectively. This value indicates a greenish or reddish color tendency in the G.CHNF film. These results are related to the presence of 1-4 linked-2-amino-2-deoxy-D-glucopyranose in CHNF (Yusof & Zain, 2019). G.ZnONPs films showed similar results, with lower a* and much higher b* values than G films, 0.18 and -0.008, respectively. Thus, the G.ZnONPs films tend to have a bluish or yellowish color. The highest decrease in L* and a* values were obtained after the addition of CHNF and ZnONPs. The addition of both has an impact on increasing the b* value of the G.CHNF.ZnONPs film.

The effect of casting thickness on the color properties of G.CHNF.ZnONPs films are shown in Table 2. The increase in casting thickness causes a decrease in the L* and a* values of the film. On the other hand, increasing casting thickness was found to increase

Comment [L36]: bold

the b* value. The tendency for the formation of reddish and dark colors inhibits the penetration of visible light through the film surface. Thus, the nutrition, color, and taste of packaged food products can be maintained (Kan, et al., 2019).

Table 1. Film color properties with variation of composition

Sample	**Parameter of Film's Color Properties**					
	L*	**a***	**b***	**YI**	**WI**	**ΔE**
G	101.25	0.38	-0.41	0.54	99.06	9.31
G.CHNF	101.04	0.26	-0.34	0.37	99.37	8.34
G.ZnONPs	99.47	0.18	-0.008	0.26	99.84	3.64
G.CHNF.ZnONPs	97.08	0.02	0.7	0.03	95.49	1.15

ΔE: total color difference, WI: whiteness indices, and YI: yellowness indices

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Table 2. Film color properties with variation of casting thickness

Sample	**Casting Thickness (mm)**	**Parameter of Film's Color Properties**					
		L*	**a***	**b***	**YI**	**WI**	**ΔE**
G. CHNF. ZnONPs	0.8	97.39	0.07	0.19	1.10	96.57	1.79
	1.0	97.26	0.05	0.21	0.07	96.22	1.80
	1.2	97.17	0.03	0.7	0.04	95.75	1.10
	1.4	97.08	0.02	0.7	0.03	95.49	1.15

Antibacterial Activity

Analysis of antibacterial activity was carried out to determine the ability of the film to inhibit contamination and bacterial growth in packaged food products. The diameter of the film inhibition zone for each bacterium is shown in Table 3-5. Film G did not show the zone of inhibition against the three test bacteria. In contrast, G.CHNF films exhibited antibacterial activity as did G.ZnONP. However, G.ZnONPs films showed much higher antibacterial activity against gram-positive bacteria than gram-negative bacteria. The presence of an outer membrane with a barrier against ROS makes gram-negative bacteria less sensitive to ROS than gram-positive bacteria (Babaei-Ghazvini, et al., 2021). With a different mechanism, the positively charged glucosamine amine group (NH_3^+) in CHNF will interact with the negatively charged outer membrane resulting in a pore that causes damage to intracellular components and causes bacterial cell death (Shankar, et al., 2017).

Comment [L38]: G film

Comment [L39]: abbreviation

G.CHNF.ZnONPs film showed the highest antibacterial activity. The synergism of CHNF and ZnONPs produces good antibacterial activity for both gram-negative and gram-positive bacteria (Amjadi, et al., 2019). The casting thickness was found to increase the antibacterial activity of the G.CHNF.ZnONP film. The increase in the availability of ROS, which results from the reaction of Zn^{2+} with oxygen, as the film thickness increases, accelerates the death of bacterial cells (Kumar, et al., 2020). The increase in thickness also causes an increase in porosity which intensifies the leakage of intracellular components leading to cell death (Kravanja, et al., 2019).

Table 3. Inhibition zone of the edible packaging components against bacteria

Component	**Inhibition Zone (mm)**		
	Staphylococcus aureus	***Escherichia coli***	***Pseudomonas aeruginosa***
CHNF	15.50	18.06	16.53
ZnONPs	19.43	17.20	16.30

Data were expressed as mean ± standard deviation (n=3)

Table 4. Antibacterial activity with a variation in composition

Sample	**Inhibition Zone (mm)**								
	Staphylococcus aureus			***Escherichia coli***			***Pseudomonas aeruginosa***		
	IZ (mm)	**K (-) (mm)**	**K (+) (mm)**	**IZ (mm)**	**K (-) (mm)**	**K (+) (mm)**	**IZ (mm)**	**K (-) (mm)**	**K (+) (mm)**
G	-			-			-		
G.CHNF	9.18	11.49	13.70	11.68	9.80	11.07	10.19	8.59	9.58
G.ZnONPs	12.65			11.38			10.03		
G.CHNF.ZnONP	14.63			12.30			11.15		

Data were expressed as mean ± standard deviation (n=3)

Table 5. Antibacterial activity of G.CHNF.ZnONP film with variations in cast thickness

Casting Thickness (mm)	**Inhibition Zone (mm)**								
	Staphylococcus aureus			***Escherichia coli***			***Pseudomonas aeruginosa***		
	IZ (mm)	**K (-) (mm)**	**K (+) (mm)**	**IZ (mm)**	**K (-) (mm)**	**K (+) (mm)**	**IZ (mm)**	**K (-) (mm)**	**K (+) (mm)**
0.8	11.15			9.66			8.38		
1.0	12.28	11.49	13.70	10.51	9.80	11.07	9.29	8.59	9.58
1.2	13.34			10.99			9.60		
1.4	14.63			12.30			11.15		

Data were expressed as mean ± standard deviation (n=3)

CONCLUSION

Gelatin-based nanocomposites containing CHNF and ZnONP have been prepared and characterized. Changes in the morphology of the G film were detected on SEM images with the addition of CHNF and ZnONP. Chitosan nanofibers create interconnected pore structures, while the presence of ZnONP suppresses the volume of free space in the gelatin matrix. As a result, the gelatin matrix's increase in density and compactness and the formation of a less permeable structure resulted in a G/CHNF/ZnONP film with the best mechanical and barrier properties. G/CHNF/ZnONP nanocomposites were also found to have the highest antibacterial activity compared to other films. With these unique features, G/CHNF/ZnONP films are the potential for packaging applications, to get better food quality and longer shelf life.

Comment [L40]: DSC and transparency test results have not been included in the conclusion

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REFERENCES

Comment [L41]: Please write references using Mendeley

Al-Tayyar, N. A., Youssef, A. M., & Al-Hindi, R. R. (2020). Antimicrobial packaging efficiency of ZnO-SiO2 nanocomposites infused into PVA/CS film for enhancing the shelf life of food products. *Food Packaging and Shelf Life, 25*. doi:https://doi.org/10.1016/j.fpsl.2020.100523

Amjadi, S., Emaminia, S., Davudian, S. H., Pourmohammad, S., Hamishehkar, H., & Roufegarinejad, L. (2019). Preparation and characterization of gelatin-based nanocomposite containing chitosan nanofiber and ZnO nanoparticles. *Carbohydrate Polymers, 216*, 376-384. doi:https://doi.org/10.1016/j.carbpol.2019.03.062

Amjadi, S., Hamishehkar, H., & Ghorbani, M. (2019). A novel smart PEGylated gelatin nanoparticle for co-delivery of doxorubicin and betanin: A strategy for enhancing the therapeutic efficacy of chemotherapy. *Materials Science and Engineering: C, 97*, 833-841. doi:https://doi.org/10.1016/j.msec.2018.12.104

ASTM. (2005). Morphological, physical, antimicrobial and release properties of ZnO nanoparticles-loaded bacterial 700 cellulose films. In *Annual book of ASTM*. Philadelphia: American Society for Testing and Materials.

Babaei-Ghazvini, A., Acharya, B., & Korber, D. R. (2021). Antimicrobial Biodegradable Food Packaging Based on Chitosan and Metal/Metal-Oxide Bio-Nanocomposites: A Review. *Polymers, 13*(16), 2790. doi:https://doi.org/10.3390/polym13162790

Bahar, A., Kusumawati, N., & Muslim, S. (2020). Preparation and characterization of goatskin gelatin as halal alternative to bovine gelatin. *RASAYAN J. Chem., 13*(1), 85-98. doi:http://dx.doi.org/10.31788/RJC.2019.1245409

Bahar, A., Rusijono, & Kusumawati, N. (2018). Extraction and characterization of the base halal gelatin on bovine bone. *Advances in Engineering Research, 171*. doi:https://doi.org/10.2991/snk-18.2018.10

Comment [L42]: subcript

Bahar, A., Rusijono, R., & Kusumawati, N. (2019). The Effect of curing and extraction time against yield and quality of type b gelatin from goat bone. *Atlantis Highlights in Chemistry and Pharmaceutical Sciences, 1*, 5-9. doi:https://doi.org/10.2991/snk-19.2019.2

Boura-Theodoridou, O., Giannakas, A., Katapodis, P., Stamatis, H., Ladavos, A., & Barkoula, N.-M. (2020). Performance of ZnO/chitosan nanocomposite films for antimicrobial packaging applications as a function of NaOH treatment and glycerol/PVOH blending. *Food Packaging and Shelf Life, 23*. doi:https://doi.org/10.1016/j.fpsl.2019.100456

Cai, L., Shi, H., Cao, A., & Jia, J. (2019). Characterization of gelatin/chitosan ploymer flms integrated with docosahexaenoic acids fabricated by diferent methods. *Scientific Reports, 9*. doi:https://doi.org/10.1038/s41598-019-44807-x

Chen, C., Deng, S., Yang, Y., Yang, D., Ye, T., & Li, D. (2018). Highly transparent chitin nanofiber/gelatin nanocomposite with enhanced mechanical properties. *Cellulose, 25*, 5063-5070. doi:https://doi.org/10.1007/s10570-018-1915-z

Chen, C., Wang, Y., Yang, Y., Pan, M., Ye, T., & Li, D. (2018). High strength gelatin-based nanocomposites reinforced by surface-deacetylated chitin nanofiber networks. *Carbohydrate Polymers, 195*, 387-392. doi:10.1016/j.carbpol.2018.04.095

Davoodi, S., Davachi, S. M., Golkhajeh, A. G., Shekarabi, A. S., & Abbaspourrad, A. (2020). Development and characterization of Salvia macrosiphon/chitosan edible films. *ACS: Sustainable Chemistry and Engineering, 8*(3), 1487-1496. doi:https://doi.org/10.1021/ascsuschemeng.9b05894

Ebrahimi, S., Fathi, M., & Kadivar, M. (2019). Production and characterization of chitosan-gelatin nanofibers by nozzle-less electrospinning and their application to enhance edible film’s properties. *Food Packaging and Shelf Life, 22*. doi:https://doi.org.10.1016/j.fpsl.2019.100387

Ge, S., Liu, Q., Li, M., Liu, J., Lu, H., Li, F., . . . Xiong, L. (2018). Enhanced mechanical properties and gelling ability of gelatin hydrogels reinforced with chitin whiskers. *Food Hydrocolloids, 75*, 1-12. doi:https://doi.org/10.1016/j.foodhyd.2017.09.023

Hari, K. D., Garcia, C. V., Shin, G.-H., & Kim, J.-T. (2021). Improvement of the uv barrier and antibacterial properties of crosslinked pectin/zinc oxide bionanocomposite films. *Polymers, 13*(15). doi:https://doi.org/10.3390/polym13152403

Kamdem, D. P., Shen, Z., Nabinejad, O., & Shu, Z. (2019). Development of biodegradable composite chitosan-based films incorporated with xylan and carvacrol for food packaging application. *Food Packaging and Shelf Life, 21*. doi:https://doi.org/10.1016/j.fpsl.2019.100344

Kan, J., Liu, J., Yong, H., Liu, Y., Qin, Y., & Liu, J. (2019). Development of active packaging based on chitosan-gelatin blend films functionalized with Chinese hawthorn (Crataegus pinnatifida) fruit extract. *International Journal of Biological Macromolecules, 140*, 384-392. doi:10.1016/j.ijbiomac.2019.08.155

Kravanja, G., Primozic, M., Knez, Z., & Leitgeb, M. (2019). Chitosan-based (nano)materials for novel biomedical applications. *Molecules*, *24*(10), 1960. doi:10.3390/molecules24101960

Kumar, S., Mudai, A., Roy, B., Basumatary, I. B., Mukherjee, A., & Dutta, J. (2020). Biodegradable hybrid nanocomposite of chitosan/gelatin and green synthesized zinc oxide nanoparticles for food packaging. *Foods, 9*(9), 1143. doi:https://doi.org/10.3390/foods9091143

Kusumawati, N., Bahar, A., Maria, M. S., & Muslim, S. (2019). Impact of curing and extraction time on yield and quality of base gelatin from goat skin. *IOP Conference Series: Earth and Environmental Science, 347*. doi:10.1088/1755-1315/347/1/012083

Lin, L., Gu, Y., & Cui, H. (2019). Moringa oil nanoparticles embedded gelatin nanofibers for food packaging against Listeria monocytogenes and Staphylococcus aureus on cheese. *Food Packaging and Shelf Life, 19*, 89-93. doi:https://doi.org/10.1016/j.fpsl.2018.12.005

Liu, C., Huang, J., Zheng, X., Liu, S., Lu, K., Tang, K., & Liu, J. (2020). Heat sealable soluble soybean polysaccharide/gelatin blend edible films for food packaging applications. *Food Packaging and Shelf Life, 24*. doi:https://doi.org/10.1016/j.fpsl.2020.100485

Nandiwilastio, N., Muchtadi, T. R., Suyatma, N. E., & Yuliani, S. (2019). Pengaruh penambahan lilin lebah dan nanopartikel seng oksida terhadap sifat fisik dan mekanis film berbasis kitosan. *Jurnal Teknologi dan Industri Pangan, 30*(2), 119-126. doi:https://doi.org/10.6066/jtip.2019.30.2.119

Comment [L43]: translate (Engglish)

Ngo, T. M., Dang, T. M., Tran, T. X., & Rachtanapun, P. (2018). Effects of zinc oxide nanoparticles on the properties of pectin/alginate edible films. *International Journal of Polymer Science, 2018*. doi:https://doi.org/10.1155/2018/5645797

Quero, F., Padila, C., Campos, V., Luengo, J., Caballero, L., Melo, F., . . . Enrione, J. (2018). Stress transfer and matrix-cohesive fracture mechanism in microfibrillated cellulose-gelatin nanocomposite films. *Carbohydrate Polymers, 195*, 89-98. doi:https://doi.org/10.1016/j.carbpol.2018.04.059

Roy, S., & Rhim, J.-W. (2021). Fabrication of bioactive binary composite film based on gelatin/chitosan incorporated with cinnamon essential oil and rutin. *Colloids and Surfaces B: Biointerfaces, 204*. doi:https://doi.org/10.1016/j.colsurfb.2021.111830

Shankar, S., Wang, L.-F., & Rhim, J.-W. (2017). Preparation and properties of carbohydrate-based composite films incorporated with CuO nanoparticles. *Carbohydrate Polymers, 169*, 264-271. doi:https://doi.org/10.1016/j.carbpol.2017.04.025

Soradech, S., Nunthanid, J., Limmatvapirat, S., & Luangtana-anan, M. (2017). Utilization of shellac and gelatin composite film for coating to extend the shelf life of banana. *Food Control, 73*, 1310-1317. doi:https://doi.org/10.1016/j.foodcont.2016.10.059

Susmitha, A., Sasikumar, K., Rajan, D., Padmakumar M, A., & Nampoothiri, K. M. (2021). Development and characterization of corn starch-gelatin based edible films incorporated with mango and pineapple for active packaging. *Food Bioscience, 41*. doi:https://doi.org/10.1016/j.fbio.2021.100977

Wafi, A., Atmaja, L., & Ni'mah, Y. L. (2020). Analisis kuat tarik dan elongasi film gelatin-kitosan. *Alchemy Journal of Chemistry, 8*(1). doi:https://doi.org/10.18860/al.v8i1.9097

Wang, H., Ding, F., Ma, L., & Zhang, Y. (2021). Edible films from chitosan-gelatin: physical properties and food packaging application. *Food Bioscience, 40*. doi:https://doi.org/10.1016/j.fbio.2020.100871

Yadav, S., Mehrotra, G. K., & Dutta, P. K. (2021). Chitosan based ZnO nanoparticles loaded gallic-acid films for active food packaging. *Food Chemistry, 334*. doi:https://doi.org/10.1016/j.foodchem.2020.127605

Yusof, N. A., & Zain, N. M. (2019). The effect of ZnO nanoparticles on the physical, mechanical, and antibacterial properties of chitosan-gelatin hydrogel films. *Jurnal Teknologi, 81*(2). doi:https://doi.org/10.11113/jt.v81.12605

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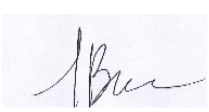

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