Development of pH-Responsive smart hydrogel incorporating basil eugenol and anthocyanin for controlled drug delivery

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Abstract
This study aimed to develop a pH-responsive smart hydrogel system based on chitosan–acrylic acid, fortified with butterfly pea (*Clitoria ternatea*) extract as a visual stability sensor and loaded with basil (*Ocimum basilicum*) leaf derived eugenol as a natural antidepressant. The hydrogel was synthesized through free-radical polymerization and characterized in terms of drug loading, release behavior, and structural interactions. Drug release tests under simulated physiological conditions revealed minimal eugenol release at pH 6.8 (oral cavity), limited release at pH 1.2 (gastric environment), and maximal release at pH 7.4 (intestinal environment). The formulation containing 2.6% acrylic acid demonstrated optimal performance, achieving high release levels in the intestine without premature loss. FTIR analysis confirmed successful grafting of acrylic acid, the incorporation of eugenol and anthocyanins, and the presence of new intermolecular interactions that enhanced structural stability. These findings indicate that the developed hydrogel holds significant potential as a controlled drug delivery system, combining therapeutic efficacy with an integrated colorimetric monitoring function.
Keywords: Smart hydrogel, chitosan–acrylic acid, eugenol, controlled drug release, anthocyanin sensor

1. Introduction

Mental health has increasingly become a critical issue, particularly in relation to the rising prevalence of depressive disorders within society. The World Health Organization (WHO) reported that 280 million people worldwide were affected by depression in 2023. Meanwhile, the Global Burden of Disease (2024) revealed that the prevalence of depression in Indonesia increased from 2.8% in 2021 to 3.8% in 2023. This situation underscores an urgent concern, as depression not only affects individual quality of life but also has significant implications for productivity and social well-being. Conventional treatments for depression typically rely on chemical-based medications, which may lead to long-term side effects such as organ damage and drug resistance. Therefore, there is a growing need to explore natural alternatives that are safer and potentially more effective (Rahma & Suzana, 2024).

Basil leaves (*Ocimum basilicum*) possess potential as a natural antidepressant agent due to their active compounds, particularly eugenol, which has been demonstrated to exert calming and synergistic effects (Tripathy e*t al.,* 2024). Eugenol is widely recognized for its therapeutic properties; however, it is highly susceptible to degradation under environmental factors such as light and temperature, as well as oxidative agents like OH and O_3 radicals present in the atmosphere (Aburel *et al*., 2021). The direct utilization of eugenol presents challenges related to stability and the effective delivery of active compounds to targeted sites within the body. To address these limitations, modern drug delivery technologies such as pH-responsive hydrogels may be employed (Yadav *et al.,* 2024). Hydrogels are hydrophilic polymers that serve as effective drug delivery systems owing to their high drug-loading capacity, ease of fabrication, and ability to provide controlled drug release (Ghosh et al., 2024). Among the biopolymers considered safe for pharmaceutical applications, chitosan is widely utilized as a base material for drug delivery systems. Chitosan, characterized by hydroxyl and amino groups, exhibits strong tissue-binding properties, thereby enhancing circulation time, therapeutic efficacy, and controlled release of drugs (Salahuddin *et al.,* 2024).

Effective hydrogels for drug delivery can be synthesized from a combination of chitosan and acrylic acid. The incorporation of anionic charges ($-COO^-$) from acrylic acid into chitosan enables the hydrogel to swell under varying pH conditions, thereby facilitating drug release at the appropriate pH while preventing premature degradation (Zhu *et al.,* 2024). In a study on hydrogel synthesis from chitosan and acrylic acid, Wang *et al.* (2017) reported a low crosslinking efficiency, with only about 0.5% of maleic anhydride (MAH) groups in chitosan-g-(maleic anhydride) (CSMAH) participating in

crosslinking with poly(acrylic acid) (PAA) chains. This low efficiency resulted in limited hydrogel stability, underscoring the need to enhance crosslinking density. Increasing crosslinking can reduce hydrogel pore size, thereby influencing drug loading capacity and release rate. Kopac *et al*. (2020) demonstrated that the average pore size of hydrogels decreased exponentially with higher concentrations of crosslinking agents, reaching a minimum at 2% concentration and a maximum at 0.25%. This challenge may be addressed by increasing the concentration of acrylic acid, which promotes the formation of larger pores, thus enhancing the diffusion rate of drug molecules and water (Sahoo & Biswal, 2024).

The novelty of this research lies in identifying an optimal balance between crosslinking efficiency and pore size, thereby enhancing the drug delivery capacity of hydrogels while simultaneously incorporating a colorimetric sensor to monitor the stability of active compounds. This sensor is derived from anthocyanins present in butterfly pea (*Clitoria ternatea*) extract, functioning as a visual indicator to detect the degradation of active compounds. Consequently, it provides crucial information regarding the stability and therapeutic effectiveness of the drug. The general objective of this research is to develop a pH-responsive smart hydrogel based on chitosan–acrylic acid, fortified with butterfly pea (*Clitoria ternatea*) extract as a drug quality sensor and loaded with basil leaf-derived antidepressant compounds. Specifically, this study aims to examine the effect of varying acrylic acid compositions on the physical characteristics of the hydrogel, including loading capacity and thickness; to investigate the influence of acrylic acid composition on the chemical properties of eugenol-loaded hydrogels, particularly crosslinking efficiency and pH responsiveness mediated by the pH-sensitive sensor; and to evaluate the impact of acrylic acid composition on the release profile of eugenol from the hydrogel as an antidepressant agent.

2. Method

The research method explains the design activities, scope of the study, objects, materials, tools, main location, data sources, techniques of data collection, definition of operational research variables, and techniques of analysis.

For Service to the Community, the methodology explained starts from stage preparation, implementation, compilation of reports and publications. In addition, the process of collaboration with partners is also explained. in a systematic way.

2.1. Synthesis of CSMAH.

A total of 0.5 g of chitosan was dissolved in 40 mL of 2.5 wt% acetic acid solution, followed by the slow addition of 2.6 g maleic anhydride dissolved in 1 mL acetone under ice-cooling conditions. The mixture was stirred at room temperature for 8 hours and subsequently precipitated in 500 mL of acetone. The solid product was washed three times with acetone and then dried under vacuum at 50 °C for 48 hours.

2.2. Anthocyanin Extraction.

A total of 100 g of butterfly pea (*Clitoria ternatea*) flowers was macerated with 1 L of an ethanol–water mixture (7:3, v/v) at 4 °C for 24 hours. The mixture was then filtered and centrifuged at 200 rpm for 10 minutes. The anthocyanin extract was stored at 4 °C until further use.

2.3. Synthesis of Dried Hydrogel.

The dried hydrogel was synthesized through free-radical polymerization using ammonium persulfate as an initiator, with CSMAH and acrylic acid serving as cross-linkers. First, 1.4 g of NaOH was dissolved in 40 mL of distilled water at room temperature, followed by the addition of 0.05 g of CSMAH and acrylic acid according to the experimental variables. The mixture was stirred until it became transparent, after which 0.01 g of ammonium persulfate and 8% (v/v) anthocyanin extract were added. The resulting hydrogel was immersed in 500 mL of a methanol–water solution (7:3, v/v) to remove unreacted residues, cut into thin cylindrical shapes, and dried in an oven at 50, 55, 60, and 65 °C until a constant weight was achieved.

2.4. Basil Leaf Extraction.

Fresh basil leaves (10 g) were extracted using ultrasonication in 100 mL of 52.48% aqueous ethanol for 7.75 minutes at 90% amplitude, with the beaker placed in an ice bath to maintain temperature. The extract was then filtered using Whatman No. 5 filter paper, and the ethanol was evaporated using a rotary evaporator.

2.5. Eugenol Analysis.

Eugenol analysis was conducted using UV–Vis spectrophotometry. Standard solutions were prepared at concentrations of 0, 5, 10, 15, 20, 25, and 35% (v/v) in 70% ethanol. The absorbance of each standard solution was measured at 304 nm to construct a calibration curve. Subsequently, the absorbance of the samples was measured at the same wavelength, and the eugenol concentration was calculated based on the calibration curve.

2.6. Drug Loading.

The dried hydrogel was immersed in 50 mL of basil leaf extract and stirred for 24 hours at 37 °C. After incubation, the hydrogel was rinsed with distilled water to remove residual surface extract. The decrease in extract concentration was measured using UV–Vis spectrophotometry at 304 nm to determine the amount of extract loaded into the hydrogel.

2.7. Drug Release Analysis.

The release of the antidepressant was evaluated in phosphate buffer solutions at pH 6.8, 1.2, and 7.4, maintained at 37 °C. The drug-loaded hydrogel was first immersed in 60 mL of pH 6.8 buffer for 1 minute, then transferred to pH 1.2 buffer for 2 hours, and subsequently placed in pH 7.4 buffer for 4 hours. The amount of antidepressant released was quantified using UV–Vis spectrophotometry at 304 nm

3. Results and Discussion

3.1. Eugenol Release Test

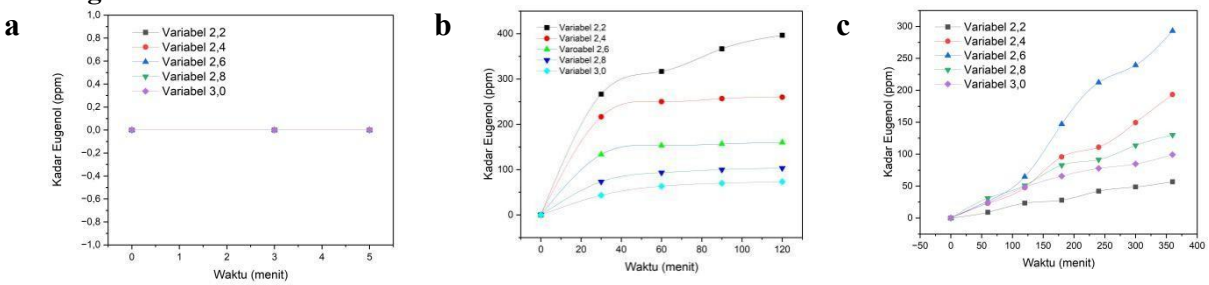

Figure 1. Eugenol release test results at pH a. 6.8; b. 1.2; c. 7.4

The release of eugenol was evaluated under simulated physiological conditions in three different media: pH 6.8 (saliva/oral cavity), pH 1.2 (stomach), and pH 7.4 (intestine). The results showed that at pH 6.8, almost no eugenol release was detected up to the 5th minute across all acrylic acid variables. This phenomenon is attributed to the very short contact time and the neutral conditions of saliva, which are insufficient to trigger hydrogel network expansion, thereby resulting in negligible eugenol diffusion. Such a condition can be considered advantageous, as it prevents premature drug loss during the initial oral contact phase, in line with the principle of pH-sensitive hydrogel-based drug delivery systems that provide site-specific release (Hong *et al.,* 2024).

Under gastric conditions (pH 1.2), the release profile exhibited a gradual increase in eugenol concentration over time; however, the extent of release strongly depended on the acrylic acid content in the formulation. Samples with lower acrylic acid concentrations (e.g., 2.2) showed the highest release, whereas higher acrylic acid levels (2.4–3.0) resulted in reduced release. This can be explained by the chemical properties of –COOH groups, which become protonated in acidic environments,

causing the hydrogel network to contract. With higher crosslinking density, the diffusion pathways for drug molecules become more restricted, thereby limiting drug release in the stomach. This pattern is consistent with previous reports indicating that acrylate-based hydrogels tend to adopt a collapsed state under low pH conditions, leading to minimal swelling and restricted drug release (Liaqat *et al.,* 2024; Suhail *et al.,* 2022).

A different behavior was observed at pH 7.4, which represents the intestinal environment. In this medium, eugenol release increased significantly due to the ionization of carboxyl groups (–COOH → –COO^-), which induced electrostatic repulsion between polymer chains. This mechanism caused the hydrogel to swell, enlarging pore structures and facilitating greater diffusion pathways for drug molecules. Interestingly, the formulation containing 2.6 acrylic acid exhibited the highest eugenol release among all variables, reaching approximately 293 ppm at 120 minutes. This composition represents the optimal point, where the number of ionized groups was sufficient to induce substantial swelling without being hindered by excessive network density. In contrast, formulations with higher acrylic acid content (2.8 and 3.0) demonstrated lower release due to overly dense crosslinking that restricted diffusion, while the lowest acrylic acid variable (2.2) was also suboptimal owing to the limited number of ionizable groups. This phenomenon is consistent with the findings of Noureen et al. (2023), which highlighted the existence of an optimal acrylate-to-polysaccharide ratio required to balance swelling capacity and drug retention.

The release study demonstrated that the developed hydrogel system exhibited well-defined pH-responsive characteristics, with minimal release in the oral cavity, limited release in the stomach, and maximal release in the intestine. Among the tested formulations, the hydrogel containing 2.6 acrylic acid was identified as the most promising for controlled intestinal delivery of eugenol-based drugs, as it achieved high release levels at the target site without significant losses during the early stages of the gastrointestinal tract.

3.2. FTIR Analysis

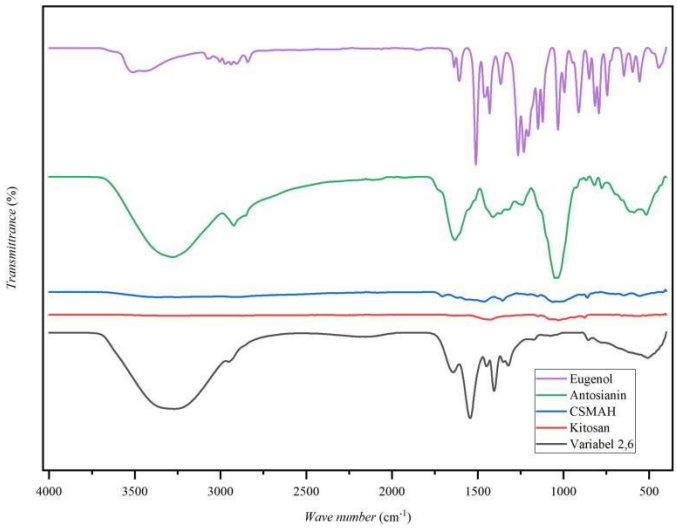

Figure 2. FTIR spectra of eugenol, anthocyanins, CSMAH, chitosan, and hydrogel formulation with 2.6% acrylic acid

The FTIR results showed the reference spectra of the pure components eugenol, anthocyanins, CSMAH, and chitosan compared with the spectrum of the 2.6 acrylic acid formulation. The eugenol spectrum was characterized by a distinctive band of phenolic –OH groups at 3200–3500 cm^{-1} and phenolic/ether C–O stretching vibrations in the range of 1260–1030 cm^{-1}. Anthocyanins exhibited conjugated C=O and aromatic C=C bands at 1700–1500 cm^{-1}, along with fingerprint C–O bands at 1200–1000 cm^{-1}. CSMAH displayed a carbonyl C=O absorption band at 1700–1730 cm^{-1}, whereas chitosan showed a broad –OH/–NH stretching band (3200–3500 cm^{-1}) and strong C–O/C–O–C stretching bands within 1200–1000 cm^{-1}.

In the 2.6 acrylic acid formulation, significant spectral changes were observed. The broadening and overlapping of the –OH/–NH stretching band indicated the formation of new hydrogen bonds

among chitosan, anthocyanins, eugenol, and the carboxylate groups of acrylate. The C=O band near 1700 cm^{-1} shifted and overlapped with the chitosan amide band (1650–1590 cm^{-1}), suggesting the presence of ionic $–NH_3^+\cdots{}^-COO^-$ interactions characteristic of acrylic acid grafting. Moreover, the characteristic bands of eugenol and anthocyanins remained detectable, although with increased intensity and slight shifts, confirming the successful incorporation of both bioactive components within the hydrogel matrix through molecular interactions.

These findings are consistent with Noureen et al. (2023), who reported that grafting in hydrogels is typically indicated by the disappearance of certain precursor bands, shifts in the C=O band position, and the emergence of new characteristic peaks. Drug loading in chitosan-based hydrogels is often reflected by the overlap of drug bands with matrix bands without the loss of the primary fingerprints (Malik et al., 2021). Similarly, Suhail et al. (2022) highlighted that the incorporation of active molecules into hydrogels can be confirmed through band overlap and minor FTIR shifts. Hong et al. (2024) further emphasized that FTIR analysis of chitosan-based hydrogels not only confirms the presence of components but also reveals intermolecular bonding changes that influence stability, swelling capacity, and drug release behavior. Collectively, these results indicate that the hydrogel system retained the characteristic fingerprints of eugenol, anthocyanins, and CSMAH, while modifications induced by acrylic acid strengthened ionic interactions within the hydrogel network, leading to a more stable structure and enabling a controlled drug release mechanism.

4. Conclusion

This study successfully developed a pH-responsive smart hydrogel system based on chitosan–acrylic acid, fortified with butterfly pea (*Clitoria ternatea*) extract as a colorimetric stability sensor and loaded with basil leaf-derived eugenol as a natural antidepressant. The hydrogel demonstrated desirable site-specific drug release behavior, with negligible release in the oral cavity, limited release in the gastric environment, and significant release in the intestinal medium. Among the tested formulations, the hydrogel containing 2.6% acrylic acid exhibited the most favorable performance, achieving high release efficiency at the intestinal target site without premature drug loss. FTIR analysis confirmed successful grafting of acrylic acid onto the chitosan backbone, the incorporation of eugenol and anthocyanins, and the formation of new intermolecular interactions that enhanced structural stability. These findings highlight the potential of the developed hydrogel as a controlled-release carrier for natural antidepressants, combining therapeutic efficacy with an integrated visual monitoring system.

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References

Aburel, O. M., Pavel, I. Z., Dănilă, M. D., Lelcu, T., Roi, A., Lighezan, R., Muntean, D. M., dan Rusu, L. C. 2021. Pleiotropic effects of eugenol: the good, the bad, and the unknown. *Oxidative Medicine and Cellular Longevity*. 20(19):1-15.

Ahmad, N. A., Heng, Y. L., Salam, F., Zaid, M. M. H., dan Hanifah, A. S. 2019. A colorimetric pH sensor based on *Clitoria* sp and *Brassica* sp for monitoring of food spoilage using chromametry. *Sensors*. 19(21):1-19.

Ali, A., dan Ahmed, S. 2024. *Polysaccharides-Based Hydrogels*. Edisi ke-1, Elsevier. India.

Chen, X., Low, H. R., Loi, X. Y., et al. (2019). Fabrication and evaluation of bacterial nanocellulose/poly(acrylic acid)/graphene oxide composite hydrogel: Characterizations and biocompatibility studies for wound dressing. *Journal of Biomedical Materials Research Part B: Applied Biomaterials, 110*(2), 2140–2151. https://doi.org/10.1002/jbm.b.34309

Ghosh, S., Kumar, N., dan Chattopadhyay, S. 2024. Electrically conductive "smart" hydrogels for on-demand drug delivery. *Asian Journal of Pharmaceutical Sciences*. 20(1):1-22.

Global Burden of Disease Study. 2024. Global Burden of Disease Report 2024. Institute for Health Metrics from https://www.healthdata.org/gbd. Diakses tanggal 10 Mei 2025.

Handayani, L., Aprilia, S., Arahman, N., dan Bilad, R. M. 2024. Identification of the anthocyanin profile from butterfly pea (*Clitoria ternatea L.*) flowers under varying extraction conditions evaluating its potential as a natural blue food colorant and its application as a colorimetric indicator. *South African Journal of Chemical Engineering*. 49:151-161.

Hong, F., Qiu, P., Wang, Y., Ren, P., Liu, J., Zhao, J., & Guo, D. (2024). Chitosan-based hydrogels: From preparation to applications—A review. *Food Chemistry: X, 21*, 101094. https://doi.org/10.1016/j.fochx.2023.101095

Ijaz, H., Tulain, U. R., Minhas, M. U., et al. (2022). Design and in vitro evaluation of pH-sensitive crosslinked chitosan-grafted acrylic acid copolymer (CS-co-AA) for targeted drug delivery. *International Journal of Polymeric Materials and Polymeric Biomaterials, 71*(6), 336–348. https://doi.org/10.1080/00914037.2020.1833011

Kopač, T., Ručigaj, A., dan Krajnc, M. 2020. The mutual effect of the crosslinker and biopolymer concentration on the desired hydrogel properties. *International Journal of Biological Macromolecules*. 159:557–569.

Koshy, R. R., Reghunadhan, A. Mary, K. S., Pillai, S. P., Joseph, S., dan Pothen, A. L. 2022. pH indicator films fabricated from soy protein isolate modified with chitin nanowhisker and *Clitoria ternatea* flower extract. *Current Research in Food Science*. 5:743-751.

Liaqat, H., Khalid, A., Riaz, T., et al. (2024). pH-Sensitive hydrogels fabricated with hyaluronic acid as a matrix for targeted mesalamine release. *ACS Omega, 9*(38), 22083–22095. https://doi.org/10.1021/acsomega.4c03240

Malik, N. S., Ahmad, M., Alqahtani, M. S., et al. (2021). β-cyclodextrin chitosan-based hydrogels with tunable pH-responsive properties for controlled release of acyclovir: Design, characterization, safety, and pharmacokinetic evaluation. *Drug Delivery, 28*(1), 1093–1108. https://doi.org/10.1080/10717544.2021.1921074

Noureen, S., Noreen, S., Ghumman, S. A., et al. (2023). A novel pH-responsive hydrogel system based on *Prunus armeniaca* gum and acrylic acid: Preparation and evaluation as a potential candidate for controlled drug delivery. *European Journal of Pharmaceutical Sciences, 189,* 106555.https://doi.org/10.1016/j.ejps.2023.106555

Pelin, I. M., Stancu, I.-C., Dinescu, S., & Costache, M. (2023). Tri-component hydrogel as template for nanocrystalline hydroxyapatite. *Gels, 9*(11), 884. https://doi.org/10.3390/gels9110905

Rahma, S.P. dan Suzana, D. 2024. Antidepressant activity test of basil leaf essential oil (*Ocimum basilicum L.)* with open field test (OFT) method. *Jurnal Farmasi dan Farmakoinformatika.* 2(1):9–22.

Sahoo, D. R., dan Biswal, T. 2024. A comparative study of the drug delivery behaviour of poly acrylic acid (PAA) reinforced with chitosan, guargum and pectin. *Materials Today*: *Proceedings*. 11(1):1-14.

Salahuddin, A., Ashraf, A., Ahmad, K., dan Hou, H. 2024. Recent advances in chitosan-based smart hydrogel for drug delivery systems. *International Journal of Biological Macromolecules*. 280(1):135803.

Silva, E. R., Teixeira, L. J. Q., Ribeiro, J. A., Oliveira, R. L., & Oliveira, F. C. (2021). The performance evaluation of Eugenol and Linalool microencapsulated by PLA on their activities against pathogenic bacteria. *Materials Today Chemistry, 21*, 100517. https://doi.org/10.1016/j.mtchem.2021.100493

Suhail, M., Liu, J. Y., Hsieh, W. C., et al. (2022). Designing of pH-responsive ketorolac tromethamine loaded hydrogels of alginic acid: Characterization, in-vitro and in-vivo evaluation.

Arabian Journal of Chemistry, 15(5), 103590. https://doi-org.proxy.undip.ac.id/10.1016/j.arabjc.2021.103590

Suryani, P. R., Zulissetiana, E. F., dan Prananjaya, B. A. 2019. Antidepressant activity of basil leaves essential oil (*Ocimum basilicum*) in male balb/c mice. *Sriwijaya International Conference on Medical and Sciences.* 26-27, Oktober 2018, Sumatra Selatan, Indonesia. 1246(012065):1-8.

Suttiarporn, P., Taithaisong, T., Namkhot, S., & Luangkamin, S. (2025). Enhanced eugenol composition in clove essential oil by deep eutectic solvent–based ultrasonic extraction and microwave-assisted hydrodistillation. *Molecules, 30*(3), 504. https://doi.org/10.3390/molecules30030504

Thang, N. H., Chien, T. B., dan Cuong, D. X. 2023. Polymer-based hydrogels applied in drug delivery: an overview. *Gels*. 9(523): 1-38.

Tripathy, N. S., Sahoo, L., Paikray, S. K., dan Dilnawaz, F. 2024. Eugenol: The phytocompound's potential biological applications. *BioNanoScience*. 14:4044–4055.

Uddin, M. S., Mahin, M. R., Rumi, M. H., & Haque, M. S. (2024). Effect of crosslinking agents on chitosan hydrogel: A review. *Gels, 10*(7), 421. https://doi.org/10.3390/gels10070421

Wang, Y., Wang, J., Yuan, Z., Han, H., Li, T., Li, L., dan Guo, X. 2017. Chitosan cross linked poly(acrylic acid) hydrogels: drug release control and mechanism. *Colloids and Surfaces B: Biointerfaces*. 152: 252-259.

Yadav, P., Singh, S., Jaiswal, S., dan Kumar, R. 2024. Synthetic and natural polymer hydrogels: a review of 3D spheroids and drug delivery. *International Journal of Biological Macromolecules*. 280(4):136126.

Zhu, H., Sun, H., Dai, J., Hao, J., dan Zhou, B. 2024. Chitosan-based hydrogels in cancer therapy: drug and gene delivery, stimuli-responsive carriers, phototherapy and immunotherapy. *International Journal of Biological Macromolecules*. 282(2):137047.