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(Research) Article

Formulation and Evaluation of Hydrogel Preparations from Jeneponto Horse Oil (Equus caballus)

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Abstract: Horse oil, traditionally used as a medicinal remedy, contains essential amino acids that contribute to the repair of bones, muscle tissues, and skin, particularly in wound healing. Hydrogel preparations serve as topical wound dressings that provide a cooling effect, reduce inflammation, and support faster wound recovery. The aim of this research was to develop and evaluate the stability of a hydrogel formulated with horse oil (Equus caballus). The study involved heating horse fat at 105°C for two hours using an oven, followed by formulating the extracted oil into a hydrogel with a sodium carboxymethyl cellulose (NaCMC) base. The preparation underwent several tests including organoleptic, homogeneity, pH, spreadability, adhesion, viscosity, and stability evaluations. Results showed that formulations F1, F2, and F3 satisfied all evaluation criteria and maintained stability without changes in physical characteristics or pH levels.

Keywords: Horse Oil; Hydrogel; Jeneponto; Stability Test; Topical preparation

1. Introduction

A wound is a disruption in the continuity of tissue that can be caused by physical injury, chemical injury, or biological factors. The wound healing process involves complex stages, including hemostasis, inflammation, tissue proliferation, remodeling, and regeneration. To support this process, local therapy using appropriate topical preparations is very important to accelerate healing and reduce the risk of infection and complications (Harliantika & Noval, 2021).

Wound healing is a complex biological process involving a series of stages, from hemostasis to tissue remodeling. In recent decades, the use of hydrogel-based topical preparations has become increasingly popular as a wound therapy medium due to their ability to maintain wound moisture and provide comfort to patients. Hydrogels can also be formulated with various active ingredients that support the tissue regeneration process and accelerate wound healing (Harliantika & Noval, 2021).

Hydrogel, as a form of modern topical preparation, has the unique ability to maintain wound moisture, which is essential in accelerating tissue healing and reducing pain. In addition, hydrogel provides a clean environment that is protected from microbial contamination. Hydrogel formulations can be enriched with natural active ingredients to enhance wound healing and reduce inflammation (Silalahi et al., 2023).

Horse oil (Equus caballus) from Jeneponto Regency is a natural ingredient that has traditionally been used in wound treatment. The bioactive compounds contained in this oil, such as essential fatty acids like linoleic acid and oleic acid, act as anti-inflammatory agents and accelerate tissue cell regeneration. (Octasari, 2021; Salenda, 2018). Previous studies indicate that horse oil can accelerate granulation and epithelialization of wounds (Nur Intang et al., 2023; Fadila, 2025).

However, efforts to develop horse oil in the form of hydrogel as a modern topical preparation are still limited. Therefore, this study aims to formulate and evaluate Jeneponto horse oil hydrogel, as well as assess the physical stability of the preparation to ensure its safety and effectiveness for use as a wound healing therapy.

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This study aims to formulate a hydrogel based on Jeneponto horse oil and test its physical and chemical stability. Stability testing is conducted to ensure that the hydrogel preparation remains effective and safe during its shelf life (Harliantika & Noval, 2021; Octasari, 2021)..

2. Preliminaries or Related Work or Literature Review

Wound healing is a complex biological process involving multiple stages, including hemostasis, inflammation, tissue proliferation, remodeling, and regeneration. Each phase plays a critical role: hemostasis stops bleeding, inflammation cleans the wound, proliferation rebuilds tissue, and remodeling strengthens the repaired area. Supporting this process with appropriate topical therapies is essential to accelerate healing, reduce infection risks, and prevent complications (Harliantika & Noval, 2021).

In recent decades, topical hydrogel preparations have gained popularity due to their ability to maintain optimal moisture in the wound environment, thus aiding faster recovery and improving patient comfort. Hydrogels can be formulated with various active ingredients that promote tissue regeneration and reduce inflammation (Harliantika & Noval, 2021; Silalahi et al., 2023). Hydrogels also provide a sterile, protective environment that minimizes microbial contamination.

Horse oil (Equus caballus) from Jeneponto is traditionally recognized for its wound healing properties. It contains bioactive compounds such as essential fatty acids (linoleic and oleic acids) that have anti-inflammatory effects and stimulate cellular regeneration. Prior research has shown that horse oil accelerates granulation tissue formation and epithelialization, key steps in wound repair (Octasari, 2021; Salenda, 2018; Nur Intang et al., 2023; Fadila, 2025).

Despite these benefits, the development of horse oil-based hydrogels as modern topical wound dressings remains limited. This study aims to formulate and evaluate the physical and chemical stability of horse oil hydrogels from Jeneponto, ensuring safety and efficacy during storage and application (Harliantika & Noval, 2021; Octasari, 2021).

3. Proposed Method

Research Design

This study used a laboratory experimental method with a true experimental design. The hydrogel formulation was developed using various concentrations of horse oil as the active ingredient, as well as a combination of optimal gel bases to obtain a stable and effective hydrogel preparation.

Tools and Materials

The tools used in this study were analytical scales, stirring rods, porcelain dishes, measuring cups, beaker glasses, ovens, aluminum foil, hot plates, droppers, spatulas, glass plates, watch glasses, pH indicators, glass objects, and viscometers.

The materials used in this study were horse oil (Equus caballus), distilled water, polyethylene glycol (PEG 400), sodium carboxymethylcellulose (Na CMC), dimethyl sulfoxide (DMSO), triethanolamine (TEA), phenoxyethanol, glycerin, and pH paper.

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Table 1. Horse Oil Hydrogel Preparation Formula (Equus caballus)

Bahan	Fungsi	F1	F2	F3
Minyak Kuda (*Equus caballus*)	Active ingredient	15%	20%	25%
Na CMC	Gelling agent	3%	3%	3%
PEG 400	Surfactant	10%	10%	10%
DMSO	Co-solvent	4%	4%	4%
Gliserin	Humectan	5%	5%	5%
TEA	Alkalizing agent	0,1%	0,1%	0,1%
Phenoxyetanol	Pengawet	0,5%	0,5%	0,5%
Aquadest	Pelarut	ad. 100%	ad. 100%	ad. 100%

4. Work Procedures

The sample used in this study was horse oil (Equus caballus) obtained from Jeneponto Regency, South Sulawesi Province. The oil was extracted from fat found in the horse's abdomen, then washed thoroughly, cut into small pieces, and finally drained so that it was ready for the next process.

Sample Preparation

The horse oil production process is carried out using a modified dry rendering method, in which horse fat is extracted at a high temperature using an oven at 105°C for 2 hours. The resulting oil is then stored in glass bottles and sealed tightly to maintain its quality.

Hydrogel Preparation

The gel base is made using Na CMC by first dissolving Na CMC in hot water. Next, ingredients such as PEG 400, dimethyl sulfoxide (DMSO), phenoxyethanol, glycerin, and triethanolamine (TEA) are slowly added and mixed until a homogeneous mixture is formed. Once the gel base has formed properly, horse oil is added gradually while continuing to stir until homogeneous. The entire homogeneous mixture is then tested and evaluated to determine the quality of the resulting hydrogel preparation.

Hydrogel Evaluation

The evaluation of the preparation was carried out through several tests to determine the physical and chemical quality of the gel preparation.

Organoleptic Test

Organoleptic testing aims to visually observe the physical properties of preparations, such as texture, aroma, and color, to ensure product uniformity and suitability (Wahidah et al., 2024).

pH Test

The pH test was conducted by weighing 1 gram of gel sample and dissolving it in 10 mL of distilled water, then measuring the pH using pH indicator paper to ensure that the preparation did not cause skin irritation. The ideal pH value according to SNI 16-3499-1996 is between 4.5 and 8 (Wahidah et al., 2024).

Spreading Power Test

The spreadability test aims to ensure that the gel spreads evenly when applied to the skin. The test is conducted by placing 1 gram of gel on a 20x20 cm glass plate, covering it with another glass plate, and then placing a 100 gram weight on top. The diameter of the spread gel is measured after one minute. According to SNI No. 06-2588, a good standard is between 5 and 7 cm (Wahidah et al., 2024).

Adhesion Test

The adhesion test was conducted by placing 0.5 grams of gel between two glass slides, pressing with a weight of 80 grams for one minute, then recording the gel release time. The gel was considered to have good adhesion if the release time was more than one second (Reinard et al., 2022).

Homogenity Test

Homogeneity testing is performed by applying the gel to a glass slide to observe whether the gel has a uniform texture without any coarse particles that can be felt (Wahidah et al., 2024).

Viskosity Test

The viscosity test was conducted by weighing 5 grams of hydrogel, placing it in a beaker, and then measuring it using a viscometer at a rotation speed of 30 rpm for 3 minutes. According to SNI, a good viscosity result ranges from 3,000 to 50,000 cP (Wahidah et al., 2024; Setiawan et al., 2018).

Stability Test

Stability testing using the cycling test method was conducted by storing the gel preparation at 27°C for 24 hours, then transferring it to an oven at 40°C for 24 hours for one cycle. The test was conducted for three cycles and observed for phase separation or syneresis in the gel (Setiawan et al., 2023).

Data Analysis

Data processing of the physical stability evaluation results of gel preparations includes organoleptic, homogeneity, pH, adhesion, spreadability, and viscosity tests, which are presented descriptively. The data obtained was then analyzed using Microsoft Excel to calculate the mean and standard deviation using the AVERAGE and STDEV formulas. For adhesion and viscosity testing, statistical analysis was performed using the one-way ANOVA method to determine significant differences between the formulas tested.

5. Result

Table 2. Organoleptic Test Before Cycling Test

Uji Organoleptic	Formulation		
	FI	**FII**	**FIII**
Color	White	White	White
Odor	Rose scent	Rose scent	Rose scent
Texture	Semisolid (Gel)	Semisolid (Gel)	Semisolid (Gel)

Table 3. Organoleptic Test After Cycling Test

Uji Organoleptic	Formulation		
	FI	**FII**	**FIII**
Color	White	White	White
Odor	Rose scent	Rose scent	Rose scent
Texture	Semisolid (Gel)	Semisolid (Gel)	Semisoli d (Gel)

Tabel 4. Homogenity Test Before Cycling Test

Formulation	Replication	Homogenitas
FI	1	Homogen
	2	Homogen
	3	Homogen
FII	1	Homogen
	2	Homogen
	3	Homogen
FIII	1	Homogen
	2	Homogen
	3	Homogen

Tabel 5. Homogenity Test After Cycling Test

Formulation	Replication	Homogenitas
FI	1	Homogen
	2	Homogen
	3	Homogen
FII	1	Homogen
	2	Homogen
	3	Homogen
FIII	1	Homogen
	2	Homogen
	3	Homogen

Tabel 6. pH Test Before Cycling Test

Formulation	Replication			Average
	1	2	3	
FI	6	6	6	6
FII	6	6	6	6
FIII	6	6	6	6

Tabel 7. pH Test After Cycling Test

Formulation	Replication			Average
	1	2	3	
FI	6	6	6	6
FII	6	6	6	6
FIII	6	6	6	6

Tabel 8. Spreading Power Test Before Cycling Test

Formulation	Replication			Average ± SD
	1	2	3	
FI	6 cm	6 cm	6 cm	6 ± 0
FII	6 cm	6 cm	6 cm	6 ± 0
FIII	6 cm	6cm	6 cm	6 ± 0

Tabel 9. Spreading Power Test After Cycling Test

Formulation	Replication			Average ± SD
	1	2	3	
FI	6 cm	6 cm	6 cm	6 ± 0
FII	6 cm	6 cm	6 cm	6 ± 0
FIII	6 cm	6 cm	6 cm	6 ± 0

Tabel 10. Adhesion Test Before Cycling Test

Formulation	Replication			Average ± SD	Sig
	1	2	3		
FI	1,91	4,74	2,59	3.5 ± 1.1	
FII	4,90	2,48	2,17	2.5 ± 0.1	0,761
FIII	2,38	2,36	2,46	2.4 ± 0.0	

Tabel 11. Adhesion Test After Cycling Test

Formulation	Replication			Average ± SD	Sig
	1	2	3		
FI	3,69	1,95	2,82	2.8 ± 0.87	
FII	3,10	1,87	2,42	2.4 ± 0.6	0,639
FIII	2,94	2,54	3,07	2.85 ± 0.2	

Tabel 12. Viscosity Test Before Cycling Test

Formulation	Viscosity (mP.S)			Average ± SD	Sig
	1	2	3		
FI	7.844	9.064	9.178	8695,333±739,4764	
FII	7.764	8.591	9.568	8641±903,0388	0,366
FIII	9.641	9.675	9.194	9503,333±268,4294	

Tabel 13. Viscosity Test After Cycling Test

Formulation	Viscositas (mP.S)			Average ± SD	Sig
	1	2	3		
FI	6.501	6.189	6.553	6414,333±196,8688	
FII	5.070	6.937	6.098	5726,333±1049,708	0,694
FIII	6.982	5.172	6.724	6292,667±979,0615	

6. Discussion

This research was conducted in the pharmaceutical technology laboratory of the Pelamonia Institute of Health Sciences in Makassar. The sample used was horse oil derived from horse fat collected in Jeneponto Regency. The fat was then cleaned and cut into small pieces to be prepared using a modified dry rendering method, whereby fat extraction was carried out at a high temperature using an oven at 105°C for two hours. The resulting oil was stored in glass bottles and sealed tightly as a preservation measure, followed by the formulation of the preparation.

Hydrogel preparations are a type of pharmaceutical preparation with a semi-solid consistency that has a hydrophilic base. One of its advantages is ease of use and cleaning after use. In addition, the intermolecular forces in hydrogels reduce molecular mobility, thereby providing good and stable viscosity in the preparation. However, the quality of hydrogels can deteriorate due to oxidation and hydrolysis processes that occur if the water content in the hydrogel is too high, so it is necessary to pay attention to the water content in the formulation to maintain product stability (Setiawan & Dewi, 2023; Sa'diyah, 2024).

The horse oil obtained was then formulated into hydrogel preparations with different concentrations, namely 15% for Formulation I, 20% for Formulation II, and 25% for Formulation III. In the manufacture of this hydrogel preparation, Na CMC was used as a gelling agent. Na CMC itself is a cellulose derivative polymer that can expand rapidly when dissolved in hot water. Its neutral properties and ability to absorb active substances well make Na CMC an effective choice. Additionally, Na CMC has advantages over carbopol, including a higher pH value due to its neutral nature, whereas carbopol is acidic. The use of Na CMC as a gelling agent also increases the spreadability of the resulting hydrogel preparation (Mikhania, 2023).

After the hydrogel preparation was completed, a series of quality evaluation tests were conducted, including organoleptic, homogeneity, pH, spreadability, adhesion, and viscosity tests. The organoleptic test aimed to assess the physical appearance of the preparation based on observations of color, texture, and aroma. The test results showed that in formulations FI, FII, and FIII, there were no changes in color, shape, or aroma after storage. Before storage, all three formulations had a white color, rose aroma, and thick consistency, and these conditions were maintained after storage (Wahidah et al., 2024).

The homogeneity test aims to ensure the uniformity of the preparation by applying the gel to a glass object and checking for the presence of coarse particles. All FI, FII, and FIII formulations showed good homogeneity results both before and after storage (Chandra & Rahmah, 2022).

pH testing was conducted to determine the acidity level so that the preparation would not cause skin irritation. The pH test results for all formulations before and after storage showed a value of 6, which is within the safe range according to SNI 16-3499-1996, namely 4.5–8 (Wahidah et al., 2024). The average pH was calculated using Microsoft Excel and a value of 6 was obtained with little variation.

The spreadability test was conducted by placing 1 gram of gel on glass and then applying a load of 100 grams. The average spreadability value for all formulations before and after storage was 6 cm, which meets the SNI No. 06-2588 standard of 5–7 cm (Wahidah et al., 2024). Data analysis shows a standard deviation of 0, indicating very little variation in values.

The adhesion test measures the time that the gel remains between two glass objects under 80 grams of pressure for 1 minute. The longer the adhesion time, the more active substance is expected to be absorbed, resulting in a more effective therapeutic effect. The test results show differences between formulas, where FIII (25% horse oil concentration) has the lowest adhesion before storage compared to FI (15%) and FII (20%). After storage, all results still meet the requirements, which is more than 1 second. The one-way ANOVA test showed no significant difference in adhesion before ($p=0.761$) and after storage ($p=0.639$) (Reinard et al., 2022; Angga, 2024).

Viscosity testing prior to storage showed a range of 7,992–9,503 cP with differences between replicates affecting adhesion. After storage, the viscosity range was 5,840–6,414 cP, which was still within the acceptable standard of 3,000–50,000 cP. One-way ANOVA analysis also showed no significant difference in viscosity between before ($p=0.366$) and after storage ($p=0.694$) (Setiawan et al., 2018).

Stability testing was conducted using a cycling test method, which stored the preparation at temperatures of 4°C and 40°C alternately for three cycles using a climatic chamber. The test results showed no physical changes such as color, texture, aroma, or pH during the test (Setiawan et al., 2023).

7. Conclusions

This study successfully formulated a horse oil-based hydrogel preparation derived from horse fat in Jeneponto Regency using a modified dry rendering method with heating at 105°C for two hours. The horse oil obtained was then used in various concentrations (15%, 20%, and 25%) with Na CMC as a gel-forming agent. The resulting hydrogel preparation exhibited stable and homogeneous physical properties, with a pH value that was safe for the skin and spreadability and adhesion that met national standards. Viscosity test results showed that the preparation had the appropriate viscosity for topical application. Stability testing using the cycling test method proved that the horse oil hydrogel did not undergo significant physical changes during storage at alternating temperatures. Thus, horse oil hydrogel from Jeneponto has the potential to be a safe, effective, and stable topical preparation for wound healing therapy.

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