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# Effects of Four Hydrocolloid Binders on Paste-Feed Stability and Behavioral Acceptance by Rice Field Eels (*Monopterus albus*)

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**ABSTRACT**

Rice field eels (*Monopterus albus*) are economically valuable, but culture development is constrained by continued reliance on wild stocks and natural feeds. This study explored how alginate, carboxymethyl cellulose (CMC), guar gum, and carrageenan, each included at 0%, 10%, 15%, or 20%, affected the water stability of paste feed. A separate behavioral-acceptance assessment compared the control with 10% alginate, 15% CMC, 15% guar gum, and 15% carrageenan. Stability observations were conducted in triplicate. The available experimental records did not establish independent animal-level units for the acceptance assessment; therefore, acceptance outcomes are reported descriptively and should not be interpreted as confirmatory treatment effects. Water addition differed among formulations (approximately 5–20 mL), creating a confounding effect between binder treatment and moisture content. Under these conditions, feeds containing 10% guar gum or 10% carrageenan remained physically intact longer than the other tested formulations. Behavioral-response counts did not provide reliable evidence of differences among acceptance treatments. These findings are exploratory and identify guar gum and carrageenan formulations for further testing in experiments that standardize moisture content and use clearly defined, randomized, independent animal or tank replicates.

## INTRODUCTION

Eel is one type of freshwater fish that has the potential to be developed today. This is because eel has a delicious taste and eel meat is widely favored by the community. In addition, eel has a high adaptability to live in murky waters and contains mud. Furthermore, eel has a high economic value which is one of Indonesia's mainstay export commodities. Indonesian people export a lot to various countries, namely the United States, India, China, Japan and Korea. The demand for paddy eels in foreign markets reaches IDR 70,000/kg. While the price for export needs in Japan receives a price of IDR 300,000/kg (1 kg/2 tails). From KKP data (2018), in West Nusa Tenggara Province, eel production reaches 737,117.43 tons/year. The market demand for eels increases every year with a selling price of 40,000-65,000/kg. However, the need for eels still depends on catching from nature (Aulya *et al*., 2023).

The increasing market demand today, farmers only rely on catching from nature so that the production of eel seeds in nature is starting to decline. Efforts to reduce the extinction of eels are carried out by cultivating the eels. Before cultivating eels, the eels must be domesticated first to prevent the extinction of the population of species that will be threatened by their survival in nature. The domestication process is a process of fish adaptation to a new environment that can affect the habits of the fish themselves, such as habits in choosing what food or feed they like (Aulya *et al*., 2023).

Natural feed for rice field eels consists of earthworms, rice field snails, snails, maggots and so on. Feeding eels does not always depend on natural feed alone, although earthworm maintenance is relatively easy and earthworms are easy to find. However, the cost of producing earthworms is relatively high and earthworms have the disadvantage that they die easily and their storage must be good and correct compared to artificial feed that is widely sold on the market. However, the provision of artificial feed can cost 40-50% of the production cost. So to attract the appetite of eels, artificial feed is made into paste feed by adding various types of binders (Aulya *et al*., 2023).

Comparison of artificial feed with natural feed that eels can consume artificial feed even though the growth value given artificial feed is relatively low. Based on previous research from research Mashuri *et al*. (2012) showed that the growth in length and absolute weight of eels from earthworm treatment was better when compared to the pellet treatment studied in clear water. The results of previous research from the study Aulya *et al.* (2023), stated that eels can consume artificial feed, however, the growth results are lower when compared to consuming natural feed. So, to attract eels to consume artificial feed, namely by making artificial feed in the form of paste.

Artificial feed in the form of paste is one alternative method that can be used for eels because paste feed can spread the pungent smell of feed. In addition, paste feed has the advantage of being soft and can spread the smell of feed into the water, so that it can stimulate the eel's appetite. However, paste feed has the disadvantage of being easily destroyed and can settle at the bottom of the pond. Therefore, this study needs to be conducted to find out how to make paste feed. In the process of making paste feed, it is necessary to add a binder as a binder that has the function of attracting the attention of eels to approach and consume the feed given. However, before giving paste feed, the binder dose and type must first be determined. The best that can provide high feed stability and is preferred by eels. So, adding binders with the right type and amount can increase food absorption quickly (Ramadan, 2014).

The binders evaluated were alginate, CMC, guar gum, and carrageenan. Previous studies indicate that hydrocolloid binders can influence feed cohesion, water stability, and palatability, although their effects depend on feed composition, processing, and inclusion level. Mudiarti *et al*. (2023) evaluated alginate in shrimp feed, whereas Wulansari et al. (2016) reported that 5% CMC improved the physical resistance of shrimp feed in water. These findings support comparative evaluation, but they do not establish an optimal binder or dose for rice field eel paste feed.

Suryaningrum (2021) discussed microbial applications for improving fish-feed ingredients. Guar gum has also been examined as a functional feed ingredient, while carrageenan has been evaluated in aquaculture feeds, including as an immunostimulant in whiteleg shrimp (Febriani *et al*., 2013). The present study therefore focused on the physical stability and behavioral acceptance of paste-feed formulations containing these binders.

## METHODS

### Time and Place

This research was conducted from May 17 to July 24, 2024 at the Fish Production and Reproduction Laboratory, Aquaculture Study Program, Faculty of Agriculture, University of Mataram for the acclimatization process. Feed stability tests were conducted at the Nutrition Laboratory, Department of Fisheries and Marine Science, Faculty of Agriculture, University of Mataram.

### Tools and Materials

The equipment used included sieves, a blender, CCTV camera, mortar and pestle, measuring cup, cable reel, camera, electric stove, pools (2 × 1 × 0.50 m and 1.93 × 0.95 × 0.50 m), containers, bowls, trays, a water-filter pump, hoses, spoons, brushes, a stopwatch, a 10-mL syringe, a digital balance, and jars measuring 10 × 8 × 4 cm.

The materials were alginate, CMC, guar gum, carrageenan, red earthworms, HI-PRO-VITE 781-1 commercial feed, tissue, and rice field eels measuring 5–10 cm.

### Research Design

The study used an exploratory experimental approach with two assessments: paste-feed stability and behavioral acceptance. Stability was evaluated in a 4 × 4 arrangement comprising four binder types (alginate, CMC, guar gum, and carrageenan) and four inclusion levels (0%, 10%, 15%, and 20%), with three repeated observations per formulation. Because the control formulation was repeated within binder labels and formulation water volumes were not standardized, the data do not constitute a fully interpretable balanced factorial experiment. Binder, dose, and binder × dose effects therefore cannot be separated confidently.

Alginate (Al), Carboxymethyl cellulose (CMC; C), Guar gum (Gu), Carrageenan (Ca), The nominal binder inclusion levels were 0%, 10%, 15%, and 20%. The stability formulations were labeled Al0–Al20, C0–C20, Gu0–Gu20, and Ca0–Ca20 and were observed three times. The subsequent behavioral-acceptance assessment compared the control, 10% alginate, 15% CMC, 15% guar gum, and 15% carrageenan. These acceptance treatments were the formulations actually tested; the study record did not document a consistent rule linking their selection to the best stability result. Consequently, the acceptance assessment is presented as a separate exploratory comparison rather than a confirmatory test of the most stable formulation.

### Research Procedures

- **Container Preparation**

For the feed-stability assessment, jars measuring 10 × 8 × 4 cm were washed, rinsed, dried, randomly positioned, and labeled by formulation. Each jar was filled with 70 mL of freshwater.

For the behavioral-acceptance assessment, a 2 × 1 × 0.50 m concrete pool was cleaned and filled with water. A filter was installed to maintain water quality, and shelters made from raffia were provided. Eels were then introduced for acclimatization. The original records do not identify the independent experimental unit, allocation sequence, or animal reuse; accordingly, the observed responses cannot be assumed to represent 30 independent replicates.

- **Test Feed Preparation**

The test feed was prepared from HI-PRO-VITE 781-1 commercial feed (31–33% crude protein), ground and sieved, and combined with 25% crushed red earthworms. Alginate, CMC, guar gum, or carrageenan was incorporated at nominal inclusion levels of 10%, 15%, or 20%; the control contained no binder. Water addition varied among formulations, from approximately 5 to 20 mL (Table 1), to obtain a moldable paste. Because moisture content was not standardized, binder treatment and water addition were confounded, and stability differences cannot be attributed to binder type or inclusion level alone.

Table 1. Feed Formulation and Pellet Preparation Procedure

| No | Treatment | Dose | Worm (g) | Pellets (g) | Binder (g) | Water (ml) | Information |
|---|---|---|---|---|---|---|---|
| 1 | Control | 0% | 1.25 | 3.75 | - | 7.5 | The pellets are mixed first with the ground worms. Then water is added, then molded using a syringe. |
| 2 | Alginate (Al) | 10% | 1.25 | 3.25 | 0.5 | 7.5 | The pellets are mixed first with the binder. After that, the crushed worms are added. |
| | | 15% | 1.25 | 3 | 0.75 | 11.2 | Then add 100°C warm water, then stir until smooth. |
| | | 20% | 1.25 | 2.75 | 1 | 15 | Next, it is formed using a syringe. |
| 3 | CMC (C) | 10% | 1.25 | 3.25 | 0.5 | 5 | The pellets are mixed first with the binder. After that, the crushed worms are added. |
| | | 15% | 1.25 | 3 | 0.75 | 7.5 | Then add 100°C warm water, then stir until smooth. |
| | | 20% | 1.25 | 2.75 | 1 | 10 | Next, it is formed using a syringe. |
| 4 | Guar gum (Gu) | 10% | 1.25 | 3.25 | 0.5 | 10 | The pellets are mixed first with the binder. After that, the crushed worms are added. |
| | | 15% | 1.25 | 3 | 0.75 | 15 | Then add 100°C warm water, then stir until smooth. |
| | | 20% | 1.25 | 2.75 | 1 | 20 | Next, it is formed using a syringe. |
| 5 | Carrageenan (Ca) | 10% | 1.25 | 3.25 | 0.5 | 10 | The pellets are mixed first with the binder. After that, the crushed worms are added. |

| No | Treatment | Dose | Worm (g) | Pellets (g) | Binder (g) | Water (ml) | Information |
|---|---|---|---|---|---|---|---|
| | | 15% | 1.25 | 3 | 0.75 | 15 | Then add 100°C warm water, then stir until smooth. |
| | | 20% | 1.25 | 2.75 | 1 | 20 | Next, it is formed using a syringe. |

- **Eel Acclimatization**

For acclimatization, eels were transferred in water-filled containers for 15–30 min to reduce temperature shock and were allowed to leave the transfer container voluntarily. They were then fasted for 3 days and monitored until no mortality occurred for seven consecutive days while being offered earthworms.

- **Feed Stability Check**

Feed stability was operationally observed in 70 mL of water. Each molded paste-feed sample was placed in a labeled jar and the jar was gently agitated every 5 min. Four visible stages were recorded: swelling; initial detachment of particles; 'broken feed,' defined for this revision as the first visible crack or separation of the molded feed into two or more pieces; and 'unformed feed,' defined as loss of the original molded shape with dispersed or collapsed material. These operational definitions clarify the recorded endpoints but should be verified against the original observation protocol before resubmission.

- **Behavioral acceptance**

Behavioral acceptance was assessed by presenting the five paste-feed treatments in the maintenance pool and recording approach, ingestion, expulsion, and refusal events. For this revision, an approach is defined as movement toward the feed without necessarily ingesting it; ingestion as taking feed into the mouth; expulsion as releasing previously mouthed feed; and refusal as approaching without ingestion or moving away. Because an eel could display more than one behavior during an observation, these categories were not mutually exclusive. The first through fifth ingestion events and elapsed time to feeding were also recorded during a 20-min observation period. The available records do not establish whether counts refer to unique eels or repeated events, so they are interpreted descriptively.

- **Water Quality**

Some parameters measured in water quality observations that need to be observed are temperature measured using a thermometer, pH measured using a pH meter, and dissolved oxygen (DO) measured using a DO meter. Measurements were carried out 3 times during the study.

- **Data Analysis**

Feed-stability observations were summarized by formulation and explored using Kruskal–Wallis and pairwise Mann–Whitney tests in the original analysis. However, the source document did not provide exact test statistics, effect sizes, confidence intervals, adjusted P values, or the raw data needed to reconstruct a two-factor analysis. The available analysis therefore cannot separate binder, dose, and binder × dose effects or verify control of multiplicity. Behavioral-acceptance counts and times are reported descriptively because independence of observations and the experimental unit were not documented.

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## RESULTS

- **Feed Stability Test Results**

The observed times to the broken-feed endpoint are shown in Figure 1. Under the formulation conditions used, including formulation-specific water volumes, 10% guar gum and 10% carrageenan remained intact longer than the other tested formulations. Because moisture addition differed among treatments and the available output lacks exact statistics, effect sizes, confidence intervals, and multiplicity-adjusted pairwise results, this pattern should be interpreted as exploratory rather than as an isolated causal effect of binder or dose.

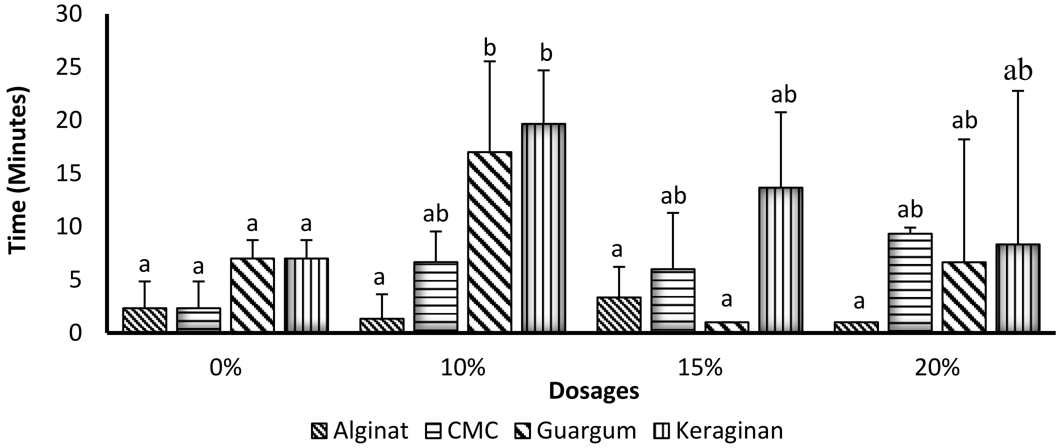


Figure 1. Time to the Broken-Feed Endpoint. Bars and Error Bars Should Be Defined from the Original Data Before Resubmission; Lowercase Letters Require Verification Against Multiplicity-Adjusted Comparisons.

The original Kruskal–Wallis analysis reported at least one difference among formulations ($P < 0.05$), but exact statistics and adjusted pairwise comparisons were not retained in the manuscript record. The apparent advantage of the 10% guar gum and 10% carrageenan formulations may reflect binder properties, moisture content, or both.

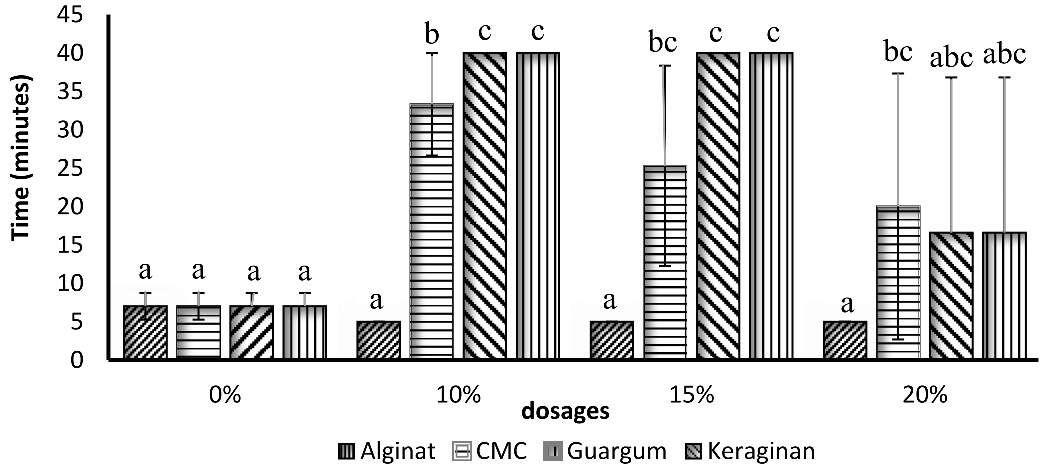


Figure 2. Time to Loss of Molded Feed Form. Bars and Error Bars Should Be Defined from the Original Data Before Resubmission; Lowercase Letters Require Verification Against Multiplicity-Adjusted Comparisons.

For loss of molded form (Figure 2), the longest observed times occurred in the 10% and 15% guar gum and carrageenan formulations. The original analysis reported P < 0.05, but the absence of exact statistics, uncertainty measures, and multiplicity control prevents a confirmatory interpretation. Water-volume differences remain an important alternative explanation.

- **Comparison of the Number of Eels Responding to Feed Given Different Paste Feeds**

A comparison of the number of eels that responded to feed with different paste feeds can be seen in Table 2.

Table 2. Number of Eels Responding to Feeding with Paste Feed

| Parameter | Number of Eels Responding to Feed (Eel) | | | | |
|---|---|---|---|---|---|
| | Control (0%) | Alginate (10%) | CMC (15%) | Guargum (15%) | Carrageenan (15%) |
| Approach events | 28 | 27 | 27 | 26 | 29 |
| Ingestion events | 12 | 13 | 17 | 9 | 8 |
| Expulsion events | 2 | 0 | 1 | 0 | 1 |
| Refusal events | 20 | 23 | 17 | 24 | 26 |

Descriptively, approach counts ranged from 26 (15% guar gum) to 29 (15% carrageenan), whereas ingestion counts ranged from 8 (15% carrageenan) to 17 (15% CMC). Because the categories could overlap and the independence and identity of observed eels were not documented, these values are behavioral counts rather than independent animal-level outcomes.

Refusal counts ranged from 17 for 15% CMC to 26 for 15% carrageenan. Expulsion was recorded one or two times for the control, 15% CMC, and 15% carrageenan formulations. These descriptive differences should not be interpreted as treatment effects without independent experimental units and a prespecified analysis.

- **Comparison of Recorded Eel Responses to Different Paste-Feed Formulations**

Comparison of the number of eel responses to different paste feeds can be seen in Table 3.

Table 3. Eel Response to Feeding Paste (Eel)

| Parameter | Number of Eel Responses to Feed (Times) | | | | |
|---|---|---|---|---|---|
| | Control (0%) | Alginate (10%) | CMC (15%) | Guargum (15%) | Carrageenan (15%) |
| First food eaten | 7 | 8 | 6 | 3 | 3 |
| Second feed eaten | 3 | 2 | 7 | 2 | 2 |
| The third feed eaten | 1 | 3 | 3 | 3 | 1 |
| The fourth feed is eaten | 1 | 0 | 0 | 1 | 2 |
| The fifth feed is eaten | 0 | 0 | 1 | 0 | 0 |

The first ingestion event was recorded most often for 10% alginate (eight observations) and least often for 15% guar gum and 15% carrageenan (three observations each). These event counts do not establish preference because repeated behavior by the same eel cannot be excluded.

The second and third ingestion events were most frequently recorded for 15% CMC (seven and three observations, respectively). Fourth and fifth events were uncommon across treatments. The findings are retained as descriptive observations only.

- **Recorded Time to Feeding (Minutes)**

Results of acceptance test of the length of time the feed is eaten (Minutes)can be seen in Figure 3.

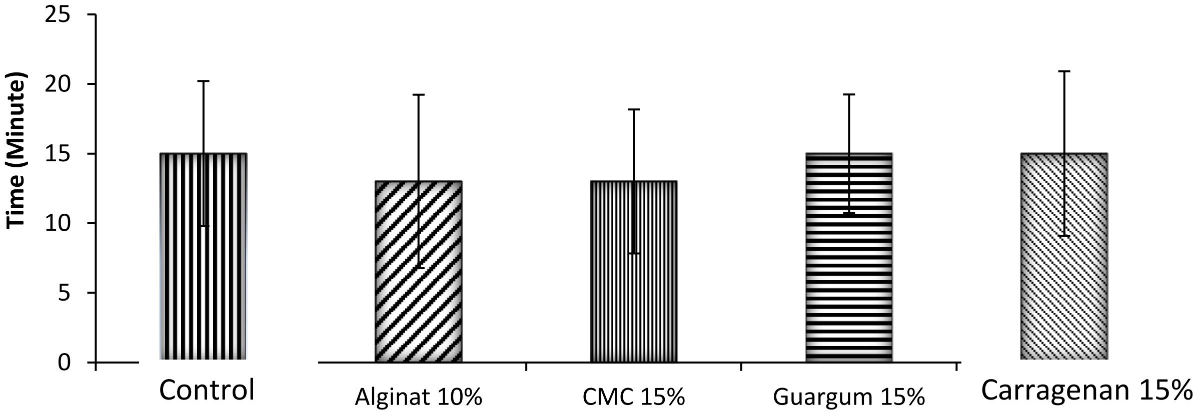


Figure 3. Recorded Time to Feeding Within the 20-min Observation Window. Bars Represent the Values Shown in the Source Figure; the Variability Measure and Sample Size Require Author Confirmation Before Resubmission.

Recorded feeding times in Figure 3 were approximately 13–15 min across treatments, not approximately 20 min as previously stated. The 20-min value refers to the observation-window duration. In the absence of verified independent units and exact inferential output, no claim of equivalence or absence of a treatment effect is made.

- **Water Quality**

The results of water quality measurements during the study can be seen in Table 3.

Table 3. Water Quality

| No | Parameter | Range Value | Optimum Value |
|---|---|---|---|
| 1 | Temperature (°C) | 27-31 °C | 27-30 ° (Santoso, 2014) |
| 2 | pH | 7.2-8 | 7-8 (Mashuri *et al*., 2012) |
| 3 | DO (mg/l) | 4.5-6 | 4-6 mg/l (Ramadhan, 2014) |

## DISCUSSION

The 10% carrageenan and 10% guar gum formulations showed the longest observed time to the broken-feed endpoint. Carrageenan and guar gum can increase cohesion and viscosity, which may contribute to physical stability (Gao *et al*., 2019). Nevertheless, water addition increased with several binder concentrations, and moisture directly affects paste viscosity, softness, and disintegration (Mulia et al., 2017, as cited in the original manuscript). The present design therefore cannot distinguish binder chemistry from formulation water content. The results identify candidate formulations for a controlled follow-up experiment; they do not demonstrate that 10% is an independently optimal dose.

For the unformed-feed endpoint, the 10% and 15% guar gum and carrageenan formulations retained their molded form longer than the control and alginate formulations.

Guar gum can function as a viscous binder and may reduce nutrient loss in water (Liu *et al*., 2022). However, physical stability is also affected by ingredient characteristics, molding, water addition, pressure, and post-processing (Yulianto, 2018). Because these factors were not fully standardized, the observed rankings are specific to the tested formulations and require confirmation under equal-moisture conditions.

The behavioral observations do not support a definitive preference ranking. Although 15% carrageenan produced the highest approach count, it also produced the lowest ingestion count and the highest refusal count. The 15% CMC treatment had the highest ingestion count, whereas 10% alginate had the greatest number of first ingestion events. These measures represent different, potentially overlapping behaviors. Prior studies have reported palatability-related responses to alginate- or CMC-containing feeds (Ahriani & Syam, 2023; Wulansari et al., 2016; Zulhisyam et al., 2020), but the present observations cannot establish causality because animal-level independence, allocation, and repeated exposure were not documented.

Figure 3 indicates feeding times of approximately 13–15 min within the 20-min observation window. Shorter latency may be compatible with greater attraction, but latency alone does not establish preference and can be influenced by prior exposure, hunger, social interaction, and repeated observation. The comparison with rapid feeding responses reported for other species (Aslamyah & Karim, 2013; Defrizal & Khalil, 2015) should therefore be interpreted cautiously.

Good water quality in the maintenance media will greatly support the growth of the fish being kept. The temperature during the study ranged between 27-31 °C. This temperature is a fairly optimal temperature in eel cultivation because according to Santoso, (2014) stated that the temperature range of 27-30 °C is the optimal temperature in eel cultivation. During the study, the pH with a range of 7.2-8 obtained was in a range that was quite optimal for rice field eel cultivation because according to Mashuri *et al*., (2012) stated that the optimum pH value in rice field eel cultivation ranged between 7-8. Dissolved oxygen (DO) during the study was 4.5-6 mg/l. The oxygen content obtained was suitable for eel maintenance.

## CONCLUSION

Under the tested formulation conditions, paste feed containing 10% guar gum or 10% carrageenan showed the longest observed physical stability in water. This result cannot be attributed solely to binder type or dose because water addition varied among formulations. The behavioral-acceptance observations did not provide confirmatory evidence of treatment differences because the independent experimental unit, animal reuse, and allocation procedures were not documented. Future work should standardize moisture content, randomize clearly defined animal- or tank-level replicates, prespecify mutually exclusive behavioral outcomes, and apply a two-factor analysis with exact statistics, effect sizes, confidence intervals, and multiplicity control.

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