

# Study on Quality Improvement Effect of Phosphate-Free Water Retainer on Squid

Huali Wang<sup>1</sup>, Feiyue Gao<sup>2\*</sup>, Xiuqin Wang<sup>1</sup>, Fang Xue<sup>1</sup>, Qingxian Zhai<sup>2</sup>

<sup>1</sup>Shandong Drug and Food Vocational College, Weihai, China, 264210

<sup>2</sup>Shandong Haidu Ocean Food Co., Ltd., Rongcheng, China, 264309

**\*Corresponding author:** Feiyue Gao, Shandong Haidu Ocean Food Co., Ltd., No. 527 Shuangshan East Road, Shidao, Rongcheng, Weihai, Shandong, China, 264309

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## Abstract

This study addresses the issues of water loss and texture deterioration during squid processing by developing a phosphate-free water retainer treatment process. Immersion concentration, time, and temperature parameters were established, and comparative experiments were conducted. Water-holding capacity, water loss rate, texture parameters, and protein structure changes were measured to analyze the regulatory effects of treatment conditions on quality. Results demonstrated that the phosphate-free system exhibited excellent performance in stabilizing water binding states and improving tissue structure. Under medium concentration conditions, samples showed the lowest centrifugal water loss and stable mass retention. Texture testing revealed reduced hardness and improved elasticity, with tissue state becoming more uniform. Electrophoresis results indicated protein transformation from high molecular weight to medium-low molecular weight, with increased exposure of hydrophilic groups and enhanced water-binding capacity. This system achieves water regulation and texture optimization without phosphate addition, demonstrating stable processing adaptability and providing support for subsequent process refinement and quality enhancement.

## Keywords

Phosphate-free water retainer; Squid; Water-holding capacity; Texture properties; Protein structure



## 无磷保水剂对鱿鱼品质改良效果研究

王华丽<sup>1</sup>、高飞跃<sup>2\*</sup>、王秀琴<sup>1</sup>、薛芳<sup>1</sup>、翟庆贤<sup>2</sup>

1 中国山东省威海市，山东药品食品职业学院，邮编 264210

2 中国山东省荣成市，山东海都海洋食品有限公司，邮编 264309

**\*通讯作者：**高飞跃，中国山东省威海市荣成市石岛双山东路 527 号，山东海都海洋食品有限公司，邮编 264309

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### 摘要

针对鱿鱼加工过程中水分流失与质构劣化问题，构建无磷保水剂处理工艺，设置浸渍浓度、时间及温度参数并开展对比实验，结合持水率、失水率、质构指标及蛋白结构变化进行检测，分析处理条件对品质的调控规律。结果表明，无磷体系在稳定水分结合状态与改善组织结构方面表现出良好效果。中等浓度条件下样品离心失水量最低且质量保持稳定，质构测试中硬度下降且弹性提升，组织状态趋于均匀，电泳结果表明蛋白由高分子向中低分子转化，亲水基团暴露增加，水分结合能力得到增强。该体系在不引入磷添加的条件下实现水分调控与质构优化，具备稳定加工适应性，可为后续工艺精细化调控与品质提升提供支撑。

### 关键词

无磷保水剂；鱿鱼；持水性；质构特性；蛋白结构

## 1 Introduction

Squid (*Illex argentinus*, *Dosidicus gigas*, etc.) is an important economic cephalopod species widely distributed in the world's major fisheries [1,2]. During low-temperature storage and processing, squid muscle is prone to myofibrillar protein denaturation, resulting in cell structure damage, free water migration, product weight loss, and texture hardening [3,4]. These quality deterioration issues seriously affect the sensory characteristics and economic value of squid products.

Traditional phosphate-containing water retainers (such as sodium tripolyphosphate, sodium hexametaphosphate) can effectively improve the water-holding capacity of aquatic products by adjusting pH value, increasing ionic strength, and promoting protein dissolution [5,6]. However, phosphate additives are subject to strict residue control in food safety standards, and excessive intake may pose health risks to consumers. In addition, phosphate-containing products may exhibit unstable quality during storage and processing [7,8]. Therefore, developing safe and efficient phosphate-free water retainers has become an important research direction in aquatic product processing [9,10].

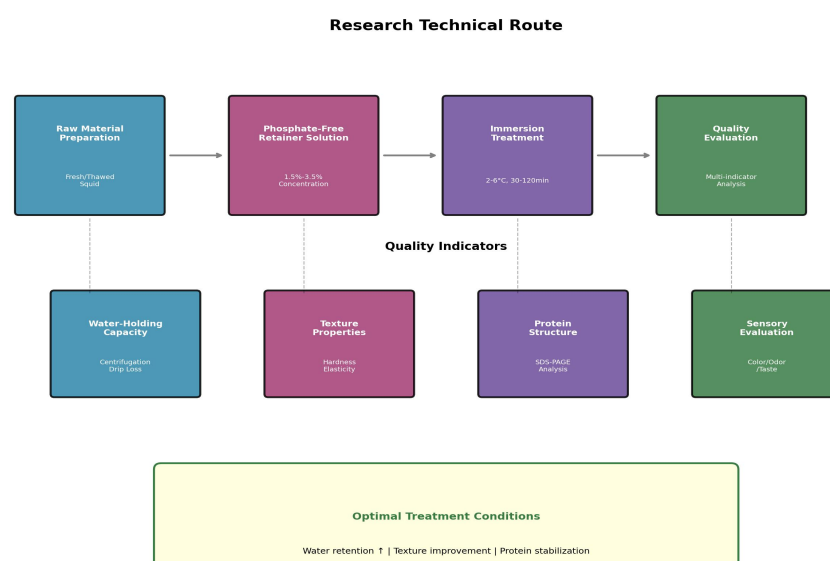
Phosphate-free water retainers are mainly composed of composite polysaccharides, organic salts, protein hydrolysates, and other natural ingredients, which achieve water retention through ion strength regulation and protein spatial structure modification [11,12]. These natural retainers not only meet food safety requirements but also possess good biocompatibility and environmental friendliness, demonstrating broad application prospects in aquatic product processing [13,14].

This study systematically investigates the application effects of phosphate-free water retainers in squid processing by constructing an immersion treatment process, establishing different treatment condition combinations, and measuring quality indicators such as water-holding capacity, texture parameters, and protein structure changes. The aim is to establish the relationship between treatment parameters and quality improvement effects, providing data support for process optimization and quality evaluation [15,16].

## 2 Materials and Methods

### 2.1 Technical Approach

The overall technical workflow of this study is illustrated in Figure 1, comprising four main stages: raw material preparation, phosphate-free retainer solution preparation, immersion treatment, and quality evaluation.



**Figure 1: Research Technical Roadmap Flowchart**

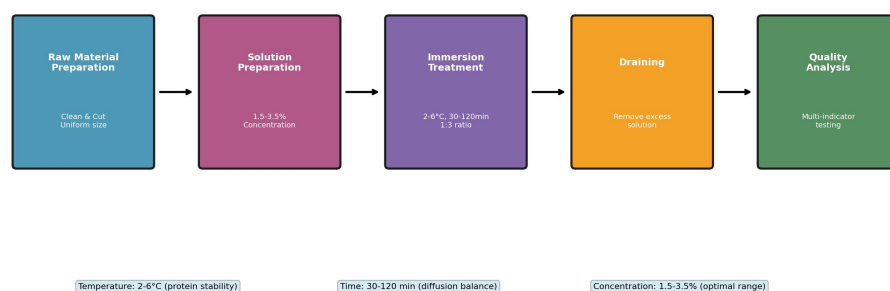
## 2.2 Raw Materials and Pre-treatment

Fresh or low-temperature thawed squid was selected as raw material, with internal organs removed and surface mucus cleaned. Samples were cut to uniform specifications, with thickness controlled to minimize mass transfer differences. Phosphate-free water retainer solutions were prepared using composite polysaccharides or organic salts as main components. Concentration was set in the range of 1.5%–3.5% with gradient configuration.

## 2.3 Immersion Treatment Process

Samples were immersed in solutions at a material-to-liquid ratio of 1:3, with temperature controlled at 2–6°C to inhibit protein degradation and maintain myofibrillar structure stability. Immersion time was set at 30–120 min for comparison. During treatment, mild agitation was maintained to enhance solution-tissue contact uniformity. The immersion process is shown in Figure 2.

**Phosphate-Free Water Retainer Immersion Process**



**Figure 2: Phosphate-Free Water Retainer Immersion Process**

## 2.4 Quality Indicators Measurement

(1) **Water-holding capacity:** After treatment, samples were subjected to surface water removal and initial mass recording. Centrifugation conditions were set at 3000 r/min for 10 min to simulate external force extrusion. Static conditions were controlled at 4°C to minimize evaporation interference. Water loss was measured and water-holding rate was calculated.

(2) **Texture analysis:** Texture testing was performed under uniform sample size and thickness conditions with consistent fiber direction. Double compression mode was used with constant compression ratio and loading speed. Hardness, elasticity, and chewiness parameters were recorded.

(3) **Protein structure analysis:** Protein extraction was performed with consistent extraction buffer system and centrifugation conditions. SDS-PAGE electrophoresis was used to analyze protein band distribution, and mass spectrometry data were combined to identify protein fragment changes. Gray value changes in each molecular weight region were recorded.

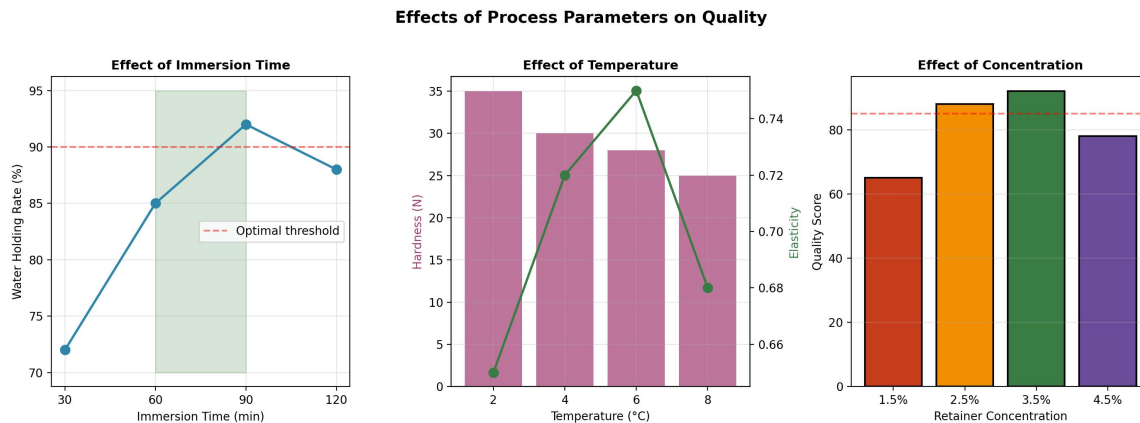
## 2.5 Statistical Analysis

All experiments were performed in triplicate. Data were expressed as mean±standard deviation and analyzed using one-way ANOVA with  $P < 0.05$  considered statistically significant.

# 3 Results and Analysis

## 3.1 Effects of Process Parameters on Quality

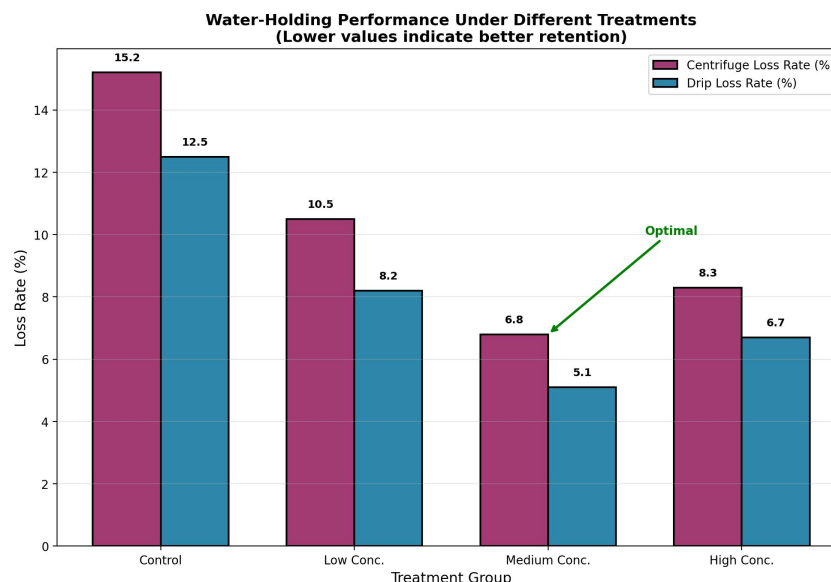
The effects of key process parameters on quality were analyzed, as shown in Figure 3. (1) Immersion time affects tissue water absorption rate and water distribution state. Extended time increases solution diffusion depth, but exceeding the critical value leads to expanded intercellular space and decreased water-holding stability. (2) Treatment temperature affects protein conformation stability and solute diffusion rate. Elevated temperature accelerates water absorption but may induce partial protein denaturation. (3) Retainer concentration determines solution osmotic pressure and ionic strength. Low concentration fails to form effective water binding, while high concentration causes tissue expansion and flavor changes.



**Figure 3: Effects of Process Parameters on Quality**

### 3.2 Water-Holding Performance Analysis

Water-holding performance test results are shown in Figure 4. Treatment group samples showed significantly reduced water loss under identical conditions, indicating altered internal water binding state in tissues. After solutes entered muscle fiber gaps and formed stable structures, water retention ability was enhanced. With increasing concentration, water-holding effect showed an initial increase followed by stabilization, indicating an optimal concentration range exists in the system.



**Figure 4: Water-Holding Performance Under Different Treatments**

Data showed that medium concentration group had the highest mass retention after centrifugation and lowest drip loss, indicating the most stable water binding degree in tissues. Control group had significantly higher water loss, reflecting the characteristic of easy water migration in untreated state. High concentration group

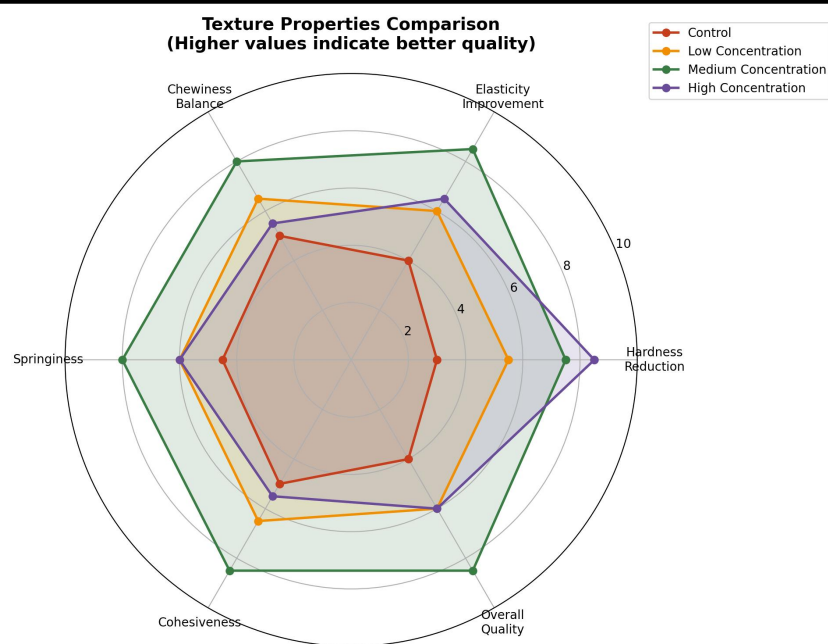
showed rebound phenomenon, indicating affected structural stability.

### 3.3 Texture Properties Analysis

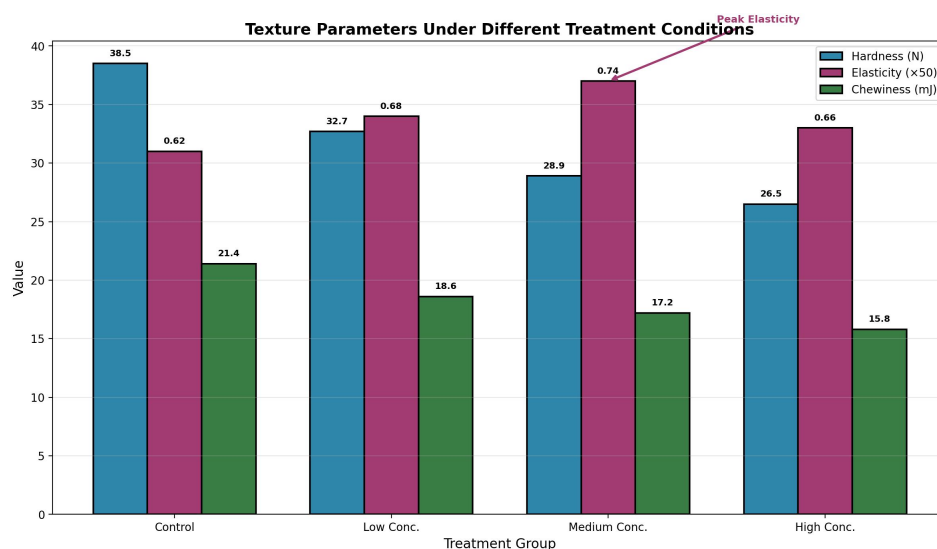
Texture parameter test results are shown in Table 1 and Figures 5-6. During treatment, phosphate-free water retainer entered myofibrillar structure and altered internal water distribution state, causing free water to transform into bound water. Myofibrillar gaps were filled with water, tissue density decreased, and stress transmission paths during force application were adjusted, manifested as reduced hardness and enhanced resilience.

**Table 1 Texture Parameters Under Different Treatment Conditions**

| Treatment Condition  | Hardness (N) | Elasticity | Chewiness (mJ) |
|----------------------|--------------|------------|----------------|
| Control Group        | 38.5         | 0.62       | 21.4           |
| Low Concentration    | 32.7         | 0.68       | 18.6           |
| Medium Concentration | 28.9         | 0.74       | 17.2           |
| High Concentration   | 26.5         | 0.66       | 15.8           |



**Figure 5: Texture Properties Comparison Radar Chart**

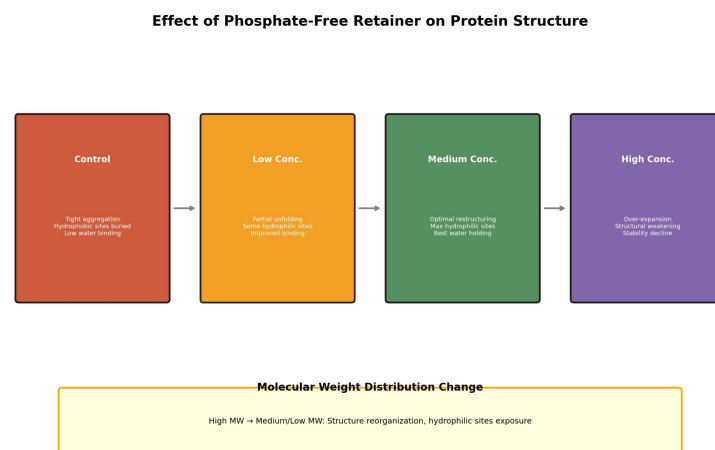


**Figure 6: Texture Parameters Under Different Treatment Conditions**

Data showed that medium concentration treatment achieved maximum elasticity with moderate hardness reduction, resulting in soft texture with resilience. Control group had higher hardness and lower elasticity, showing dense tissue. High concentration conditions showed decreased elasticity, reflecting weakened structural support capacity.

### 3.4 Protein Structure Changes

After phosphate-free water retainer entered the muscle system, it altered the ionic environment and adjusted intermolecular protein interactions, as shown in Figure 7. Partial myofibrillar proteins transformed from tight aggregation to relatively loose structures, intermolecular binding forces weakened, originally buried water in structures was redistributed, hydrophilic group exposure increased, and water binding sites increased accordingly.

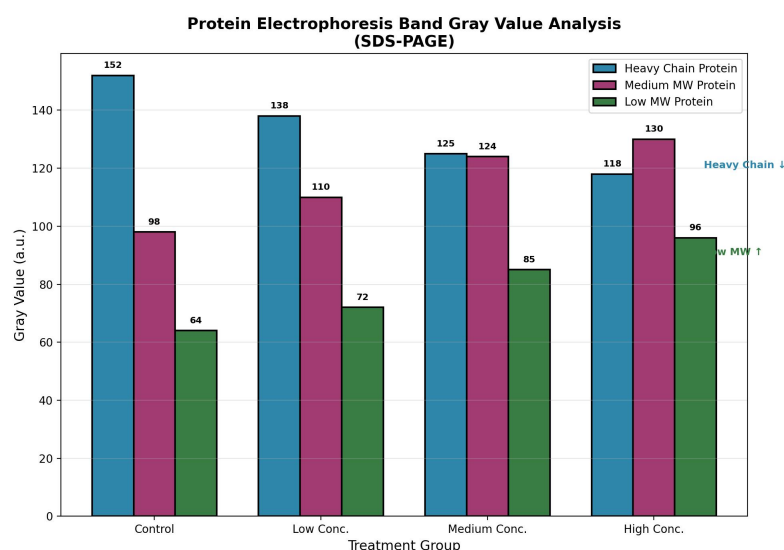


**Figure 7: Effect of Phosphate-Free Retainer on Protein Structure**

SDS-PAGE electrophoresis results are shown in Table 2 and Figure 8. When treatment intensity increased, proteins transferred from high molecular weight structures to medium-low molecular weight structures, band distribution gradually changed from concentrated to dispersed, and structural reorganization degree enhanced.

**Table 2 Protein Electrophoresis Band Gray Value Data**

| Treatment Condition  | Heavy Chain | Medium MW | Low MW |
|----------------------|-------------|-----------|--------|
| Control Group        | 152         | 98        | 64     |
| Low Concentration    | 138         | 110       | 72     |
| Medium Concentration | 125         | 124       | 85     |
| High Concentration   | 118         | 130       | 96     |

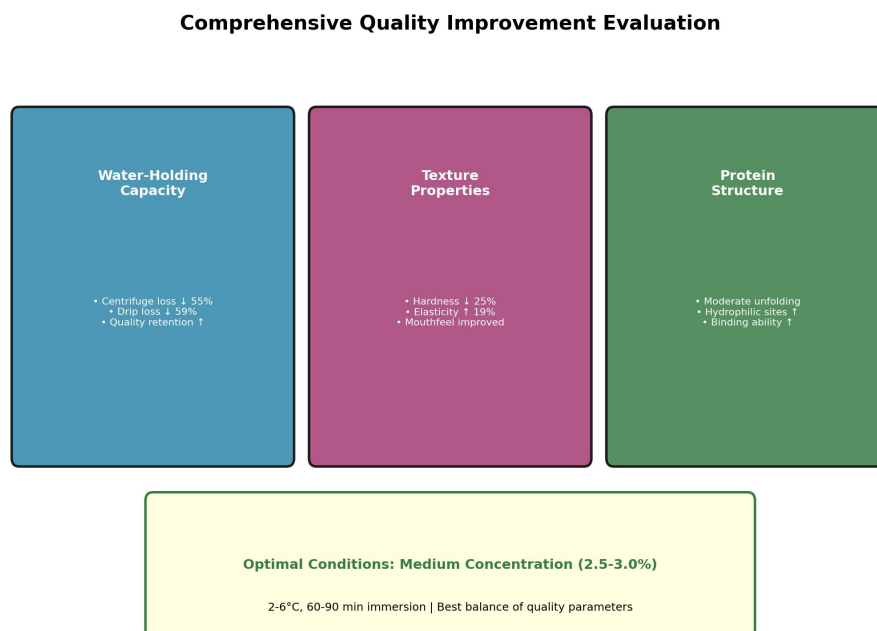


### Figure 8: Protein Electrophoresis Band Gray Value Analysis

Gray value changes showed gradual decrease in heavy chain protein and increase in medium and low molecular weight proteins, indicating protein structure reorganization. Under medium concentration conditions, structural adjustment was relatively balanced, enhancing water binding ability while maintaining tissue stability. Under high concentration conditions, excessive small molecule proportion reflected decreased structural integrity.

## 4 Discussion

This study systematically investigated the quality improvement effects of phosphate-free water retainers on squid. Results demonstrated that the phosphate-free system achieved significant improvements in water-holding capacity, texture properties, and protein structure, as shown in Figure 9. The mechanism of action is illustrated in Figure 10.



### Figure 9: Comprehensive Quality Improvement Evaluation

The water-holding mechanism of phosphate-free retainers involves multiple aspects: (1) Ionic strength regulation: Adjusting protein charge state, reducing aggregation, enhancing solubility; (2) Protein structure modification: Promoting moderate unfolding, exposing hydrophilic sites, increasing water binding sites; (3) Water binding enhancement: Facilitating free water to bound water transformation, forming hydration layers, reducing water mobility.

### Mechanism of Phosphate-Free Water Retainer Action



**Figure 10: Mechanism of Phosphate-Free Water Retainer Action**

Compared with traditional phosphate-containing retainers, phosphate-free systems have the following advantages: **(1) Safety:** No phosphate residue, meeting food safety requirements; **(2) Stability:** Quality remains stable during storage and processing; **(3) Environmental friendliness:** Reducing phosphorus emissions, environmentally sustainable; **(4) Versatility:** Can be combined with other functional ingredients to achieve comprehensive quality improvement.

This study has certain limitations: First, only short-term quality effects were evaluated, and long-term storage stability requires further verification; Second, sensory evaluation data need more comprehensive validation through larger sample testing; Third, specific molecular mechanisms of protein-retainer interactions need further investigation.

## 5 Conclusion

This study constructed a phosphate-free water retainer immersion treatment process, with concentration, time, and temperature parameters controlled in stages. Systematic measurements of water-holding capacity, texture indicators, and protein structure changes were conducted. Results showed that: (1) Under medium concentration conditions, samples exhibited the lowest centrifugal water loss and stable mass retention; (2) Texture testing revealed reduced hardness and improved elasticity, with tissue state becoming more uniform; (3) Electrophoresis results indicated protein transformation from high molecular weight to medium-low molecular weight, with increased hydrophilic group exposure and enhanced water-binding capacity; (4) Parameter deviation from the optimal range resulted in structural looseness or stability decline, indicating the process needs to operate within reasonable limits. This system achieves water regulation and texture optimization without phosphate addition, demonstrating stable processing adaptability and can replace traditional water retention treatment methods.

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