Ministry of Education and Science of Ukraine

Sumy National Agrarian University

Qualifying scientific work on the rights of the manuscript

NAN HAIJUAN

УДК 637.54:365.82

DISSERTATION

DEVELOPMENT OF LOW-FAT MEAT PRODUCTS TECHNOLOGY USING EDIBLE MUSHROOMS

Speciality: 181 – Food Technology

Field of knowledge 18 – Production and Technologies

Submitted for a scientific degree of Doctor of Philosophy

The dissertation contains the results of own research. The use of ideas, results and texts of other authors have references to the relevant source

NAN HAIJUAN ________________Nan Haijuan

NAN HAIJUAN

Scientific supervisor: Stepanova Tetiana,

PhD, Associate Professor

Sumy 2023

АНОТАЦІЯ

Нан Хайцзюнь. Розробка технології м’ясних продуктів зі зниженим вмістом жиру з використанням їстівних грибів. – Кваліфікаційна наукова праця на правах рукопису. Дисертація на здобуття ступеня доктора філософії за спеціальністю 181 – «Харчові технології» – Сумський національний аграрний університет, Суми, 2023.

Дисертаційну роботу присвячено науковому обґрунтуванню та розробці технології м’ясних продуктів зі зниженим вмістом жиру – курячих сосисок із шампіньйоном двоспоровим (*Agaricus bisporus* (Ab)).

Науково доведено доцільність використання порошку Ab як замінника жиру при виробництві нежирної курячої продукції. Було теоретично та практично обґрунтовано інноваційну стратегію розробки нової продукції з курки зі зниженим вмістом жиру із Ab.

М'ясо містить різноманітні корисні для здоров’я людини незамінні компоненти. Разом із тим, продукти з переробленого м’яса містять до 30 % жиру. Численними дослідженнями встановлено зв'язок між різноманітними хронічними захворюваннями та споживанням рафінованих м’ясних продуктів з високим вмістом жиру. Технологічні (вологоутримувальна здатність, теплові втрати, стабільність емульсії, консистенція) та органолептичні (смак, запах, консистенція, соковитість, загальна сприйнятність) властивості продукту знижуються внаслідок зниження вмісту жиру. Це впливає на потенціал ринку та споживче сприйняття м’ясних продуктів зі зниженим вмістом жиру. Тому створення м’ясних продуктів зі зниженим вмістом жиру є викликом для м’ясної галузі.

Поширений культивований сорт грибів Ab має низький вміст жиру та не володіє високою калорійністю, проте вміщує істотну кількість білку вуглеводів і харчових волокон. В роботі зазначено на значний потенціал білків, вуглеводів і харчових волокон як замінників жиру. Тому гриб Ab є ідеальною заміною тваринного жиру в м'ясних продуктах.

На початку досліджень формується наукова гіпотеза за результатами аналізу

сучасних літературних джерел, яка науково підтверджена в теоретичному аналізі та експериментальній частині. Як результат, дослідження перевіряються практичним застосуванням.

Під час експериментальних досліджень використовувалися різноманітні методи аналізу, зокрема, фізико-хімічні, мікробіологічні, біохімічні, спектральні, фізико-механічні, сенсорні, системного аналізу та комплексні, а також методи планування експерименту. Встановлено вплив Ab на основні характеристики курячого фаршу та курячих сосисок.

На підставі отриманих результатів досліджень розроблено технологію сосисок зі зниженим вмістом жиру із використанням як основної сировини – курки та Ab, а також перевірено практичним застосуванням наукову обґрунтованість результатів досліджень.

В першому розділі дисертації представлено результати аналізу літературного огляду щодо сучасних трендів споживання м’яса і м’ясної продукції, характеристики охолоджених м’ясних продуктів і прогресивні технології зниження жиру в них, застосування грибів у харчових продуктах і перспективи застосування шампіньйона двоспорового в продукції з птиці зі зниженим вмістом жиру.

Другий розділ дисертації включає план теоретичних та експериментальних досліджень, а також детальний опис матеріалів і методів дослідження, що застосовувались у роботі. Експериментальна частина була виконана в лабораторних умовах коледжу харчових наук та технології провінції Хенань, КНР. Розроблена продукція була апробована та впроваджена у виробництво двома харчовими компаніями провінції Хенань, КНР.

У третьому розділі наведено результати дослідження впливу різної кількості внесення, способу внесення та розміру частинок Ab на якість курячого фаршу, а також механізм взаємодії між Ab і курячим фаршем.

У дослідженні використовували курячу грудку як сировину та порошок Ab для заміни 2%, 4%, 6%, 8%, 10%, 20%, 30%, 40% і 50% тваринного жиру в курячому фарші відповідно. Результати показали, що в результаті заміни 20% жиру на порошок Ab значно покращились структурні показники курячого фаршу, при цьому мікроструктура стала більш компактною та однорідною. З метою більш прийнятного способу заміни жиру порошком Ab було обрано воду та соєву олію , що є найбільш споживаною в усьому світі. Результати показали, що комбінація порошку Ab і соєвої олії виявилася кращою, завдяки чому частина вільної води в курячому фарші перетворилася на зв'язану, а вихід, вологоутримувальна здатність, текстура та реологічні властивості курячого фаршу значно (P <0,05) покращились.

При використанні соєвої олії та порошку Ab із різним розміром частинок для заміни 60 % жиру результати колеляційного аналізу показали, що розмір частинок Ab корелює з виходом при тепловій обробці, водоутримувальною здатністю, значеннями L*, a*, b* і значенням pH курячого фаршу. Також розмір частинок Ab мав помірну кореляцію з пружністю. Було встановлено оптимальний розмір частинок порошку Ab D2 (100μm).

Під час додавання до розчину курячого міофібрилярного білка 1%, 2%, 4% і 6% порошку Ab відповідно, текстура гелю міофібрилярного білка покращувалася при додаванні порошку Ab в діапазоні 1% – 4%. Крім того, Ab заповнював сітку гелю та сприяв розгортанню α-спіральної структури міофібрил та утворенню β-ланцюгів міофібрилярних білків під час їх термічної денатурації, що призвело до щільної агрегованої сітчастої структури.

У четвертому розділі представлено рецептуру курячих сосисок зі зниженим вмістом жиру, з додаванням Ab, проведено їх оцінку якості, харчової цінності та показників зберігання.

Рецептурна композиція була отримана на підставі сенсорної оцінки внесення картопляного крохмалю, курячого фаршу, солі та Ab до курячих сосисок зі зниженим вмістом жиру. Було також досліджено їх хімічний склад, вміст амінокислот і жирних кислот, вихід і мікроструктуру.

Для визначення показників зміни якості курячих сосисок зі зниженим вмістом жиру під час зберігання, визначали зміни кольору, консистенції, антиоксидантних властивостей, pH, а також мікробіологічні показники сосисок протягом усього зберігання. Результати показали, що курячі сосиски зі зниженим вмістом жиру мають довший термін зберігання та більш стабільні показники якості, мінімальний термін зберігання яких становить 25 днів. За результатами досліджень було розроблено технологічну схему виробництва курячих сосисок зі зниженим вмістом жиру.

У п’ятому розділі оцінено економічну ефективність при виробництві курячих сосисок зі зниженим вмістом жиру. Результати показали, що їх промислове виробництво із використанням Ab має суттєвий економічний потенціал, переваги та заслуговує уваги при просуванні на ринок.

Таким чином, було визначено кількість, спосіб внесення та розмір частинок Ab, кількість курячого фаршу, картопляного крохмалю, солі та сполуки Ab і соєвої олії шляхом оцінки якості гелю, реологічних показників, розподілу води, мікроструктури, сенсорних характеристики, показників харчової цінності та зберігання. Успішно розроблено якісні, поживні, низькокалорійні, корисні курячі сосиски. В той же час механізм покращення властивостей курячого гелю за допомогою Ab було вивчено при взаємодії між Ab і курячим міофібрилярним білком, що забезпечило теоретичну основу для застосування Ab у м’ясних продуктах.

Ключові слова: культивовані гриби, *Agaricus bisporus*, якість м'яса курки, сосиски, жир, напівфабрикати, знижена температура, гель, технологія, білок, зберігання.

СПИСОК ПУБЛІКАЦІЙ ЗДОБУВАЧА ЗА ТЕМОЮ ДИСЕРТАЦІЇ

Публікації, що відображають основні наукові результати дисертації

Статті

1. Haijaun Nan, Kondratiuk Natalia, Stepanova Tetiana, Afanasiiev Oleksander, Sytnyk Kateryna, Suprunenko Kateryna. Vegetable protein blend technology for vegetarian sausages. Науковий вісник Полтавського університету економіки і торгівлі, 2019, 1(91), 38-46. http://doi.org/10.37734/2518-7171-2019-1-5. Стаття у фаховому виданні України. *Внесок: збір експериментальних даних.*

2. Haijaun Nan, Kondratiuk Natalia, Stepanova Tetiana, Suprunenko Kateryna Prospects of cultivated mushrooms use in technology of sausages. Bulletin of the National Technical University «KhPI» Series: New solutions in modern technologies, 2019, 2, 75-80. https://doi.org/10.20998/2413-4295.2019.02.11. Стаття у фаховому виданні України. *Внесок здобувача: збір експериментальних даних.*

3. Haijuan Nan, Bo Li, Stepanova Tetiana. Relevance of cultivated mushrooms usage in food and perspectives in sausage processing. Academic notes of V.I. Vernadskyi TNU. Series: Technical sciences, 2021, 32(71), 142-147. https://doi.org/10.32838/2663-5941/2021.2-2/22. Стаття у фаховому виданні України. *Внесок здобувача: збір експериментальних даних.*

4. Haijuan Nan, Stepanova Tetiana, Bo Li, Kondratiuk Natalia. Effect of *Agaricus bisporus* on gel properties and microstructure of chicken batters. Journal of Hygienic Engineering and Design, 2021, (9):170-178. Стаття у міжнародній наукометричній базі Scopus Q_4. *Внесок здобувача: визначення експериментальних методів і отримання експериментальних даних*

5. Haijuan Nan, Bo Li, Kondratiuk Natalia, Sylchuk Tetiana, Stepanova Tetiana. Effect of different particle sizes of *Agaricus bisporus* and soybean oil on rheological properties, moisture distribution and microstructure of chicken batters. Journal of Chemistry and Technologies, 2021, 29(2), 342-352. https://doi.org/10.15421/jchemtech.v29i2.228820. Стаття у міжнародній наукометричній базі Scopus Q_4. *Внесок здобувача: збір та отримання*

експериментальних даних, написання статті.

6. Haijuan Nan, Stepanova Tetiana, Kondratiuk Natalia, Yuanyang Nie, Bo Li. Effects of *Agaricus bisporus* on gel properties of chicken myofibrillar protein. International Journal of Food Science and Technology, 2022, 7. https://doi.org/10.1111/ijfs.15898. Стаття у міжнародних наукометричних базах Scopus Q_1 і Web of Science. *Внесок здобувача: отримання та аналіз експериментальних даних щодо впливу Agaricus bisporus на властивості гелю курячого міофібрилярного білка*

7. Haijuan Nan, Haoyu Zhou, Bo Li, Stepanova Tetiana, Kondratiuk Natalia. Effects of *Agaricus bisporus* alone or in combination with soybean oil or water as fat substitutes on gel properties, rheology, water distribution, and microstructure of chicken batters. Food Science and Technology, 2022, 42, e116121. https://doi.org/10.1590/fst.116121. Стаття у міжнародних наукометричних базах Scopus Q_2 і Web of Science. *Внесок здобувача: отримання та аналіз експериментальних даних щодо властивостей гелів, реологічних показників, розподілу води та мікроструктури курячого фаршу.*

8. Haijuan Nan, Stepanova Tetiana, Bo Li. Effects of button mushroom *Agaricus bisporus* (Agaricomycetes) and soybean oil on storage characteristics of chicken sausage. International Journal of Medicinal Mushrooms, 2023, 7. https://doi.org/10.1615/IntJMedMushrooms.2023049470. Стаття у міжнародних наукометричних базах Scopus Q_3 і Web of Science. *Внесок здобувача: визначення експериментальних методів і отримання експериментальних даних.*

Тези доповідей

9. Haijuan Nan, Stepanova Tetiana, Kondratjuk Nataliia, Bo Li (2020). Application of fat substitute in gelatinous conditioning meat products // Materials of the scientific and practical III International conference of students and postgraduates “Modern technologies of food processing”. (Dnipro, 14-15 May 2020).

10. Haijuan Nan, Bo Li, Kondratiuk Nataliia, Stepanova Tetiana (2021). Rheological properties, moisture distribution and microstructure of chicken batters with soybean oil and different particle sizes of *Agaricus bisporus* // Materials of the scientific

and practical conference "Informational and innovative technologies in hotel and restaurant business, tourism and design". (Dnipro – Opole, Poland, 1-2 December, 2021).

11. Haijuan Nan, Stepanova Tetiana, Bo Li, Kondratiuk Natalia (2021). Effect of *Agaricus bisporus* on gel properties and microstructure of chicken batters // Materials of the Food Quality and Safety, Health and Nutrition Congress. (Ohrid, Macedonia 9-11 June, 2021).

12. Haijuan Nan, Stepanova Tetiana, Kondratjuk Nataliia, Bo Li (2022). Comparison of the biological value of edible fungi and chicken meat // Materials of the International Scientific Internet Conference "The light of scientific research". (Poland 29-30 September, 2022).

13. Stepanova Tetiana, Haijuan Nan, Bo Li (2022). Modern aspects of cultivated mushrooms use in the technology of sausages // Materials of the IXTH international session of young scientific staff. (Poznań, Poland, 19-20 May, 2022).

14. Stepanova T.M., Haijuan Nan (2023). Application of fat substitute in gelatinous conditioning meat products // Materials of the first international scientific and practical conference "Sustainable food chain and security through science, knowledge and business", Kharkiv. DBTU, 2023. p. 81-82. 2023. (Kharkiv, 18 May, 2023).

Монографія

15. Modern achievements of food science in aspects of cultivated mushrooms application in food technology/Nan Haijuan, Stepanova Tetiana, Li Bo, Kondratjuk Nataliya. Monograph. - Henan Institute of Science and Technology, Xinxiang, China, Sumy National Agrarian University, Sumy, Ukraine, School of Food Science, OLDI+. 2022.

ANNOTATION

Nan Haijuan. Development of low-fat meat products technology using edible mushrooms - Qualified scientific research work manuscript.

Dissertation for the degree of Doctor of Philosophy in the speciality 181 - «Food technology» - Sumy National Agrarian University, Sumy, 2023.

This thesis is devoted to the scientific justification of the low-fat chicken sausage technology based on the low-fat chicken batters with *Agaricus bisporus* (Ab) mushroom.

This thesis presents the results of the current trend analysis of meat and meat product consumption, the characteristics of low-temperature meat products and the progress of fat-reduction technology, the application of mushrooms in food, and the application prospect of Ab in low-fat chicken products.

It was proved scientifically that the feasibility of using Ab powder as a fat substitute in the production system of low-fat chicken products.

It was identified that an innovative strategy for the development of a new low-fat chicken with Ab by theory and practice.

A variety of essential nutrients for human health can be found in meat. However, some processed meat products have up to a 30 % fat content. A significant number of research have discovered a link between various chronic diseases and consumption of high-fat refined meat products. The technical (water holding capacity, cooking losses, emulsion stability, texture, etc.) and organoleptic (taste, smell, mouthfeel, juiciness, overall acceptability, etc.) quality of the product is reduced as a result of the reduction in fat content, which has an impact on the market potential and consumer acceptance of low-fat meat products. As a result, creating low-fat meat products is a challenge for the meat business.

A widely cultivated mushroom variety, *Agaricus bisporus* (Ab), is low in fat and energy, but high in protein, carbohydrates, and dietary fiber. The potential of protein, carbohydrates and dietary fiber as fat substitutes has been demonstrated. Therefore, Ab mushroom is an ideal substitute for animal fat in meat products.

The study begins with the formation of a scientific hypothesis based on the results

of the analysis of modern literature sources, which is scientifically confirmed in theoretical analysis and experimental studies. Finally, the results of the study are verified by practical application.

During the experimental research, the methods of analysis, physicochemical, microbiological, biochemical, spectral, physical mechanics, sensory, systematic analysis and comprehensive methods and planning experimental work were used. The effects of *Agaricus bisporus* on the main characteristics of chicken batters and chicken sausage were established.

Based on the research materials and the results obtained, a technology for the production of low-fat sausages using *Agaricus bisporus* and chicken as the main raw materials was developed and the scientific validity of the research results was verified by practical application.

The first section of the thesis analyzed the trend of meat and meat product consumption, the characteristics of low-temperature meat products and the progress of fat-reduction technology, the application of mushrooms in food, and the application prospect of Ab in low-fat chicken products.

The second section includes a description of the research material, a list of the research methods used in the work and their detailed description, and a general plan for conducting theoretical and experimental studies. The experimental part was carried out mainly under experimental conditions at the College of Food Science and Technology of Henan Province, China. The developed products was approved and applied for implementation in two food companies in Henan Province, China.

The third section describes the results of the study of the effects of different addition amounts, addition methods and particle size of *Agaricus bisporus* on the quality of chicken batters, and the mechanism of interaction between *Agaricus bisporus* and chicken batters.

The study took chicken breast as raw material and Ab powder as a fat substitute to replace 2%, 4%, 6%, 8%, 10%, 20%, 30%, 40% and 50% of animal fat in chicken batters respectively. The findings showed that substituting 20% of the fat with *Agaricus bisporus* powder considerably enhanced the textural qualities of the chicken batters and made the microstructure more compact and consistent. To find a suitable way of replacing fat with *Agaricus bisporus* powder, the most economical water and the most widely consumed soybean oil worldwide were chosen to replace fat in combination with Ab powder. The results showed that the combination of Ab powder and soybean oil performed better, which made a part of free water of the chicken batters transformed into immobilized water and CY, WHC, texture, and rheological properties of the chicken batters significantly ($P < 0.05$) improved.

When soybean oil and *Agaricus bisporus* powder with different particle sizes were used to replace fat of 60 %, the correlation analysis showed that the particle size of Ab was highly correlated with the cooking yield, water holding capacity, L*, a*, b*value and pH value of chicken batters. In the meantime, the particle size of Ab had a moderate correlation with springiness. The optimal particle size of Ab powder was D2 (100μm).

When the chicken myofibrillar protein (MP) solution was added with 1%, 2%, 4% and 6% *Agaricus bisporus* powder fraction, respectively, the texture of the MP gel improved when 1% – 4% Ab powder was added. Furthermore, Ab filled in the gel network and promoted the unfolding of α-helix structure of MP and the formation of β-sheet of MP during the thermal denaturation of MP, leading to a dense aggregated network structure.

In the fourth section, the formulation of low-fat chicken sausage containing *Agaricus bisporus* was optimized, and the quality, nutritional value and storage characteristics of low-fat chicken sausage were evaluated.

The optimal formulation was obtained by sensory evaluation for the addition of potato starch, chicken essence, refined salt and *Agaricus bisporus* in low-fat chicken sausage.

Low-fat chicken sausage production, nutritional value, and quality characteristics were examined. The research assessed the chemical composition, amino acid and fatty acid content, cooking yield, and microstructure of the sausages. The findings demonstrated that low-fat chicken sausage's cooking yield was unaffected by its increased nutritional value and more logical nutritional composition.

In order to obtain the quality change pattern of low-fat sausages during storage, the changes in color, texture, antioxidant properties, pH, and total viable counts of sausages throughout storage were determined. The results showed that low-fat chicken sausages had longer shelf life and more stable storage qualities, with a minimum shelf life of 25 days. Based on the outcomes of the experiments, the technological frameworks for the manufacturing of low-fat chicken sausages was developed.

In the fifth section, the economic benefit of producing low-fat chicken sausage was evaluated. The results indicated that the industrial production of low-fat chicken sausages using *Agaricus bisporus* had good economic prospect, huge benefits, and was worth promoting.

In a word, the addition amount of Ab, addition method of Ab, the particle size of Ab, the addition amount of chicken essence, potato starch, refined salt, and the compound of *Agaricus bisporus* and soybean oil were optimized by evaluating the quality of gel, rheology, water distribution, microstructure, sensory characteristics, nutritional characteristics and storage characteristics. A high-quality, nutritious, low-calorie, healthy, low-fat chicken sausage was successfully developed. Meanwhile, the mechanism of improving chicken gel properties by *Agaricus bisporus* was discussed through the study of the interaction between *Agaricus bisporus* and chicken myofibrillar protein, which provided a theoretical basis for the application of *Agaricus bisporus* in meat products.

Keywords: cultivated mushrooms, *Agaricus bisporus*, quality of chicken meat, sausage, fat, semi-finished product, low-temperature, gel, technology, protein, storage.

LIST OF PUBLISHED WORKS ON THE THEME OF THE DISSERTATION

Articles

1. Haijaun Nan, Kondratiuk Natalia, Stepanova Tetiana, Afanasiiev Oleksander, Sytnyk Kateryna, Suprunenko Kateryna. Vegetable protein blend technology for vegetarian sausages. Науковий вісник Полтавського університету економіки і торгівлі, 2019, 1(91), 38-46. http://doi.org/10.37734/2518-7171-2019-1-5. An article in a professional publication of Ukraine. *Author's contribution: collation of experimental data.*

2. Haijaun Nan, Kondratiuk Natalia, Stepanova Tetiana, Suprunenko Kateryna Prospects of cultivated mushrooms use in technology of sausages. Bulletin of the National Technical University «KhPI» Series: New solutions in modern technologies, 2019, 2, 75-80. https://doi.org/10.20998/2413-4295.2019.02.11. An article in a professional publication of Ukraine. *Author's contribution: collation of experimental data.*

3. Haijuan Nan, Bo Li, Stepanova Tetiana. Relevance of cultivated mushrooms usage in food and perspectives in sausage processing. Academic notes of V.I. Vernadskyi TNU. Series: Technical sciences, 2021, 32(71), 142-147. https://doi.org/10.32838/2663-5941/2021.2-2/22. An article in a professional publication of Ukraine. *Author's contribution: collation of experimental data.*

4. Haijuan Nan, Stepanova Tetiana, Bo Li, Kondratiuk Natalia. Effect of *Agaricus bisporus* on gel properties and microstructure of chicken batters. Journal of Hygienic Engineering and Design, 2021, (9):170-178. An article in the international database Scopus Q_4. *Author's contribution: determination of experimental methods and acquisition of experimental data.*

5. Haijuan Nan, Bo Li, Kondratiuk Natalia, Sylchuk Tetiana, Stepanova Tetiana. Effect of different particle sizes of *Agaricus bisporus* and soybean oil on rheological properties, moisture distribution and microstructure of chicken batters. Journal of Chemistry and Technologies, 2021, 29(2), 342-352. https://doi.org/10.15421/jchemtech.v29i2.228820. Стаття у міжнародній наукометричній базі Scopus Q_4. *Author's contribution: acquisition and collation of experimental data, writing of articles.*

6. Haijuan Nan, Stepanova Tetiana, Kondratiuk Natalia, Yuanyang Nie, Bo Li. Effects of *Agaricus bisporus* on gel properties of chicken myofibrillar protein. International Journal of Food Science and Technology, 2022, 7. https://doi.org/10.1111/ijfs.15898. An article in the international databases of Scopus Q_1 i Web of Science. *Author's contribution: experimental data obtained and analyzed on the Effects of Agaricus bisporus on gel properties of chicken myofibrillar protein.*

7. Haijuan Nan, Haoyu Zhou, Bo Li, Stepanova Tetiana, Kondratiuk Natalia. Effects of *Agaricus bisporus* alone or in combination with soybean oil or water as fat substitutes on gel properties, rheology, water distribution, and microstructure of chicken batters. Food Science and Technology, 2022, 42, e116121. https://doi.org/10.1590/fst.116121. An article in the international databases of Scopus Q_2 i Web of Science. *Author's contribution: experimental data obtained and analyzed on gel properties, rheology, water distribution, and microstructure of chicken batters.*

8. Haijuan Nan, Stepanova Tetiana, Bo Li. Effects of button mushroom *Agaricus bisporus* (Agaricomycetes) and soybean oil on storage characteristics of chicken sausage. International Journal of Medicinal Mushrooms, 2023, 7. https://doi.org/10.1615/IntJMedMushrooms.2023049470. An article in the international databases of Scopus Q_3 i Web of Science. *Author's contribution: determination of experimental methods and acquisition of experimental data.*

Conference Articles or abstracts

9. Haijuan Nan, Stepanova Tetiana, Kondratjuk Nataliia, Bo Li (2020). Application of fat substitute in gelatinous conditioning meat products // Materials of the scientific and practical III International conference of students and postgraduates “Modern technologies of food processing”, (Dnipro, 14-15 May 2020).

10. Haijuan Nan, Bo Li, Kondratiuk Nataliia, Stepanova Tetiana (2021). Rheological properties, moisture distribution and microstructure of chicken batters with soybean oil and different particle sizes of *Agaricus bisporus* // Materials of the scientific and practical conference “Informational and innovative technologies in hotel and restaurant business, tourism and design”. (Dnipro – Opole, Poland, 1-2 December, 2021).

11. Haijuan Nan, Stepanova Tetiana, Bo Li, Kondratiuk Natalia (2021). Effect of

Agaricus bisporus on gel properties and microstructure of chicken batters // Materials of the Food Quality and Safety, Health and Nutrition Congress. (Ohrid, Macedonia 9-11 June, 2021).

12. Haijuan Nan, Stepanova Tetiana, Kondratjuk Nataliia, Bo Li (2022). Comparison of the biological value of edible fungi and chicken meat // Materials of the International Scientific Internet Conference "The light of scientific research". (Poland 29-30 September, 2022).

13. Stepanova Tetiana, Haijuan Nan, Bo Li (2022). Modern aspects of cultivated mushrooms use in the technology of sausages // Materials of the IX^{TH} international session of young scientific staff. (Poznań, Poland, 19-20 May, 2022).

14. Stepanova T.M., Haijuan Nan (2023). Application of fat substitute in gelatinous conditioning meat products // Materials of the first international scientific and practical conference "Sustainable food chain and security through science, knowledge and business", Kharkiv. DBTU, 2023. p. 81-82. 2023. (Kharkiv, 18 May, 2023).

Monograph

15. Modern achievements of food science in aspects of cultivated mushrooms application in food technology/Nan Haijuan, Stepanova Tetiana, Li Bo, Kondratjuk Nataliya. Monograph. - Henan Institute of Science and Technology, Xinxiang, China, Sumy National Agrarian University, Sumy, Ukraine, School of Food Science, OLDI+. 2022.

TABLE OF CONTENTS

LIST OF ABBREVIATIONS AND TERMS

Ab – *Agaricus bisporus*

CY – Cooking yield

FTIR – Fourier transform infrared

G'– The storage modulus

G″– Loss modulus

LF-NMR – Low-field nuclear magnetic resonance

MP – Myofibrillar protein

PDCAAS – Protein Digestibility Corrected Amino Acids Score

SEM – Scanning electron microscopy

TBARS – Thiobarbituric acid reactive substance

TPA – Textural profile analysis

TVC – Total viable counts

WHC – Water holding capacity

INTRODUCTION

Ukraine and China have traditional friendly relations. The cooperation between the two countries in the field of science and technology is one of the priority areas of bilateral relations, and is constantly strengthening. The two countries signed an agreement on mutual recognition of educational and scientific degrees.

Relevance of topics. Meat contains many essential nutrients for human body, such as protein, iron, zinc, selenium, vitamins, etc. Protein from animals has high biological value, and contains many essential nutritional amino acids for human body. Therefore, meat intake is beneficial. The fat content of some processed meat products is as high as 20 - 30%, which is very popular in all regions. However, a large number of studies have found that intake of high-fat refined meat products is associated with cardiovascular disease, hypertension, diabetes and other diseases. If we directly reduce the consumption of various processed meat products, it will certainly affect the sustainability and economy of the meat industry. Therefore, a more active way to solve this problem is to develop innovative processed meat products, which not only provide necessary nutrition, but also do not affect the normal health of consumers.

Consumers are also now preferring meat products that contain ingredients that are beneficial to health or do not contain ingredients that might cause health problems, which indicate a potential market opportunity for meat products that will see huge growth over the next decade. The first step in developing this healthy meat product is to reduce the high fat content of most processed meat products. However, the reduction in fat content leads to a reduction in the quality of the product in terms of technical (water holding capacity, cooking losses, emulsion stability, texture, etc.) and organoleptic (taste, smell, mouthfeel, juiciness, overall acceptability, etc.), thus affecting the marketability and consumer acceptance of low-fat meat products. Therefore, the meat industry is facing the problem of developing low-fat meat products.

Agaricus bisporus (Ab) is a cultivated mushroom variety worldwide, which is rich in nutrition and delicious taste and deeply loved by consumers. However, Ab mushroom is very easy to spoil and deteriorate, and the shelf life is only a few days, which restricts the development of Ab mushroom to a certain extent. Deep processing is an effective way to solve this problem. Ab is rich in protein, carbohydrates and dietary fiber, but low in fat and energy. The protein in Ab mushrooms has amphiphilic properties, which helps to form a stable emulsified meat emulsion system. When the polysaccharide in Ab mushrooms fully absorbs water, it can simulate the smooth taste of fat based on the physical properties of the water-like liquid system, act as a fat-like emulsification, and improve the water retention of the product. The insoluble dietary fiber in Ab mushrooms can be used as a filler, which is distributed in the gel network structure of the meat batters to improve the gel strength of the meat batters. Therefore, Ab mushroom is an ideal substitute for animal fat in meat products.

However, how to develop low-fat meat products successfully with Ab mushroom is not easy, because it involves issues such as technology, formula, nutrition and product acceptance. Therefore, establishing the action mechanism of Ab mushroom on the meat gel system, optimizing the production process parameters of low-fat meat products with Ab mushroom, and studying the nutritional and storage characteristics of low-fat meat products are the main contents of this study.

Relationship with science programs, plans, themes. The thesis work was carried out according to the main directions of Sumy National Agrarian University and Henan Institute of Science and Technology. This work was supported by the 2021 Zhongyuan science and technology innovation leading talent project (No:224200510019), 2022 Henan Province Key Science and Technology Research Project (No:222102110179)-“Development of low-fat chicken sausage with *Agaricus bisporus* and study of its mechanism of action”.

The purpose and tasks of the research. The purpose of this dissertation is to develop a low-fat chicken product containing Ab mushroom. To achieve this goal, the following tasks must be solved:

The formation rule of the low-fat chicken gel system containing Ab mushroom was established, and the process parameters of the system were determined.

The process and formula composition of the low-fat chicken batters and low-fat chicken sausage containing Ab mushroom were determined.

The feasibility of applying Ab mushroom to low-fat chicken gel system was proved theoretically.

The nutrition, edible characteristics and storage period of low-fat chicken sausages containing Ab mushroom were studied.

Analyzed the economic benefits of practical implementation of low-fat chicken sausages containing Ab mushroom. A standards document on the production processing and operating key points of low-fat chicken sausages containing Ab was developed and implemented in companies to gain industry recognition of the study results.

The object of the study - Low-fat chicken product technology using Ab mushroom as a fat substitute.

The subject of the study - Low-fat chicken gel and model system using Ab mushroom as a fat substitute.

Research methods: analysis, physicochemical, microbiological, biochemical, spectral, physical mechanics, sensory, systematic analysis and comprehensive methods, planning experimental work.

Scientific novelty of the obtained results. The interaction mechanism between Ab mushroom and chicken myofibrillar protein was systematically studied for the first time, which provided a theoretical basis for the application of Ab mushroom in meat products.

The effects of different addition amounts, addition methods and particle sizes of Ab mushroom on the gel properties, rheological properties, moisture distribution and microstructure of chicken batters were systematically studied for the first time. This is not only the first theoretical analysis of the gelation process of low-fat chicken batters containing Ab mushroom during thermal processing, but also the law of cooking yield of low-fat chicken batters containing Ab mushroom is explained from the state of water in the gel. The microstructure explained the effect of Ab mushroom on the texture of chicken batters. The results of this study confirmed the effectiveness of Ab mushroom in the low-fat chicken gel system and the feasibility of producing low-fat chicken products using Ab mushroom.

The technical parameters of the application of Ab mushroom in low-fat chicken batters and low-fat chicken sausages were determined for the first time, which can provide a reference for the industrial production of low-fat chicken products with Ab mushroom.

Low-fat chicken sausages were produced using Ab mushroom and soybean oil complex fat substitutes for the first time.

The effect of Ab mushroom and soybean oil compound fat substitutes on the nutritional quality of chicken sausage was determined for the first time, and the use of Ab mushroom to produce low-fat, nutritious and safe chicken products has been scientifically confirmed.

For the first time, The changes of oxidation, texture, color, pH value and microbiological parameters of low-fat chicken sausage containing Ab mushroom during storage were established, and the storage conditions and preservation characteristics of the new product were scientifically confirmed for the first time.

Practical significance of the obtained results. The technology of low-fat chicken batters and low-fat chicken sausage based on Ab mushroom and soybean oil as fat substitutes was developed. The developed technology improved the nutrition value and edible safety of chicken products, which can be widely used in the production of chicken products. Such as using this technology to produce low-fat chicken balls, low-fat lunch chicken meat and other emulsified chicken products.

Personal contribution of the acquirer. The research problems were analysed and scientifically demonstrated, and the research subjects, objectives and plans were formulated. Experimental parts of the research were carried out with the direct participation of the acquirer. Standard documents on the production processing and operating key points of low-fat chicken sausages containing Ab have been developed and implemented in enterprises. The author summarized the research results, drew the research conclusion, and prepared the article and thesis for publication. Acquirer's individual contributions were substantiated by regulatory and scientific literature publications.

Approbation of research results. The main research results of the dissertation work were reported, discussed and received a positive evaluation at the annual scientific conferences of professors, teaching staff, and postgraduate students of the Poznan University of Technology (Poland, 2022) and Henan University of science and technology (2019-2022). The main research results of the dissertation work were reported, discussed and received a positive evaluation at some conferences, such as "Informational and innovative technologies in hotel and restaurant business, tourism and design. December 1-2, 2021, Dnipro - Opole (Ukraine - Poland)". "Food Quality and Safety, Health and Nutrition Congress. 9-11 June, 2021, Ohrid, Macedonia". "International Scientific Internet Conference. 2022, September 29-30". "IXTH international session of young scientific staff. 2022, May 19-20. Poznań, Poland". "First international scientific and practical conference, 2023. May 18. Kharkov, Ukraine".

Publications. The dissertation materials have been published in full. The main materials of the dissertation work are presented in 15 scientific works, including: 8 articles, 1 monograph of Ukraine, 6 materials of conferences and abstracts of reports. In which, 4 articles in approved scientific specialized publications of Ukraine (1 of them in publications included in the international scientometric databases Scopus), 2 articles published in the scientific periodical of Macedonia and United States respectively, which included in the international scientometric databases Scopus, 2 articles included in the international scientometric databases Web of Science.

Dissertation structure. The dissertation consists of annotation, an introduction, five chapters, conclusions, a list of used literature sources from 207 names, including 200 foreign ones, as well as 3 appendices. The full volume of the dissertation is 161 pages of the main text, and contains 37 tables and 41 figures.

SECTION 1 LITERATURE REVIEW

1.1 Analysis of nutrition and consumption trend of meat and meat products

Meat is a concentrated nutrient source and plays an extremely important role in human growth and development, especially in the development of the brain and intelligence[1].

1.1.1 Meat nutritional composition

Meat is rich in protein of high biological value and micronutrients such as sulfur, sodium, iron, and zinc. The nutritional compositions of several common types of meat are shown in Table 1.1.

（1） Meat protein content and protein value

As seen in Table 1.1, there is considerable variation in the protein content of various meats. According to China's food composition table data[2], the average protein content of meat is 22%, with chicken breast having the highest protein content (24.6%), and pork and duck meat has the lowest protein content (15.1%, 15.5%, respectively). Meat has an amino acid score (PDCAAS) of 0.92 compared to egg whites and casein with a Protein Digestibility Corrected Amino Acids Score (PDCAAS) of 1.00 and is much higher than pinto beans, lentils, peas and chickpeas, which are widely considered important sources of protein in vegetarian meals beans (score of 0.57 - 0.71), and wheat gluten (score of 0.25) [3].

Amino acids are the main units of protein, and insufficient intake can lead to protein malnutrition. Meat protein is unique in that it is rich in all essential amino acids without limiting amino acids. Grains such as rice and wheat are especially low in lysine, while legumes are very low in methionine, so vegetarians must combine grains and legumes to get all the essential amino acids[4].

Table 1.1– Nutritional composition of several types of meat (per 100 grams of edible portion) [2]

Meat	Energy value (kcal)	Protein (g)	Fat (g)	P (mg)	Na (mg)	Fe (mg)	Zn (mg)
Chicken, average	145	20.3	6.7	166	62.8	1.8	1.46
Chicken breast	118	24.6	1.9	170	44.8	1	0.26

Chicken, leg	146	20.2	7.2	271	73.6	1.8	1.11
Chicken, wing	202	19	11.5	94	50.8	0.9	0.42
Beef, average	160	20	8.7	182	64.1	1.8	4.7
Beef, tenderloin	107	22.2	0.9	241	75.1	4.4	6.92
Beef, front-leg	105	19.2	1.8	181	69.9	2.8	4.5
Beef, back-leg	106	20.9	2	210	45.4	3.3	4.07
Pork, average	331	15.1	30.1	121	56.8	1.3	1.78
Pork, lean	143	20.3	6.2	189	57.5	3	2.99
Pork, fat	807	2.4	88.6	18	19.5	1	0.69
Pork, leg	190	17.9	12.8	185	63	0.9	2.18
Mutton, average,	139	18.5	6.5	161	89.9	3.9	3.52
Duck, average	240	15.5	19.7	122	69	2.2	1.33
Geese	251	17.9	19.9	144	58.8	3.9	1.36

（2） Fat content

As shown in Table 1.1, the fat content of different meats varies widely. Among them, pork is the fattest meat, with a fat content ranging from 6.2% to 88.6%. Beef is the leanest meat, with a fat content ranging from 0.9% to 8.7%, of which tenderloin is the leanest part of beef, with a fat content of only 0.9%. Among poultry products, chicken is the leanest meat, but the fat content of different parts of chicken varies greatly. Among them, chicken breast is the leanest part of the chicken, with a fat content of only 1.9%. Fats in meat are mainly various fatty acids and triglycerides. There are also small amounts of lecithin, cholesterol, free fatty acids and fat-soluble vitamins. Meat fat can provide more calories for the body. For decades, dietary guidelines have recommended avoiding saturated fatty acids to prevent cardiovascular disease [5]. Therefore, from a health point of view, one should intake more meat products with low-fat content, especially chicken breast.

(3) Minerals

Meat is one of the best sources of sodium, phosphorus, iron and zinc. Every mineral element has an important physiological function. As shown in Table 1.1, beef and mutton are relatively rich in mineral elements. Among them, the content of phosphorus, sodium, iron and zinc in beef tenderloin is much higher than that of other meats. This may be related to their long-term consumption of plant-based feed.

Meat is also a major source of other micronutrients vital to the human body, such as niacin, vitamins A, B_{12}, and retinol, which have important physiological activities in the human body. For example, vitamin A is an essential vitamin for human growth and development.

1.1.2 Current market and development trend of meat and meat products consumption

(1) Current market of meat consumption in the world and China

China is one of the largest meat-producing and consuming countries in the world. In the past five years, the consumption structure of the three major types of meat in the world and China is shown in Figure1.1 and Figure1.2. It can be seen that the three major meat consumption trends in the world and China are consistent, with a decrease in pork consumption and an increase in chicken consumption. From 2018 to 2019, due to the impact of African swine fever, the attributes of chicken as an alternative product became prominent, resulting in some pork consumption shifted to chicken consumption, and the growth rate of chicken consumption was significant. It also can be seen from the figure1.1 that the global consumption ratio of pork and chicken is about 1:1, and the proportion of pork consumption in China is about three times that of chicken consumption in the same period, therefore, there is still a lot of room for growth in chicken consumption. As can be seen from Figure 1.3, in the meat consumption structure, global poultry meat accounts for 40%, and China accounts for less than 30%. China's poultry meat consumption is dominated by chicken (about 63%), followed by a duck (about 29%), and goose meat ranks third (nearly 7%), which is mainly influenced by traditional Chinese food culture. The nutritional advantages of chicken with three lows and one high (low fat, low energy, low cholesterol, and high protein) can meet the growing concern of Chinese residents for a healthy diet and promote an increase in chicken consumption. It has been found that reducing the intake of red meat and meat products will reduce the risk of cancers such as rectal and breast cancer [6]. At the same time, China's population growth is still a factor that can not be ignored in driving the growth of food consumption, and its pulling effect is generally greater than the growth of individual demand based on per capita income or economic growth. Therefore, chicken consumption will continue to grow in the long term.

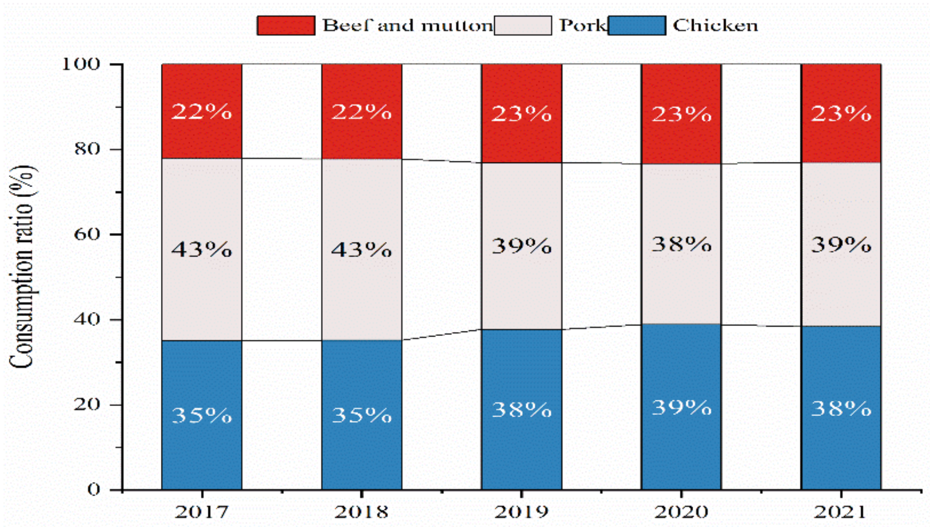

Figure 1.1– Consumption structure of the world's three major types of meat in 2017-2021

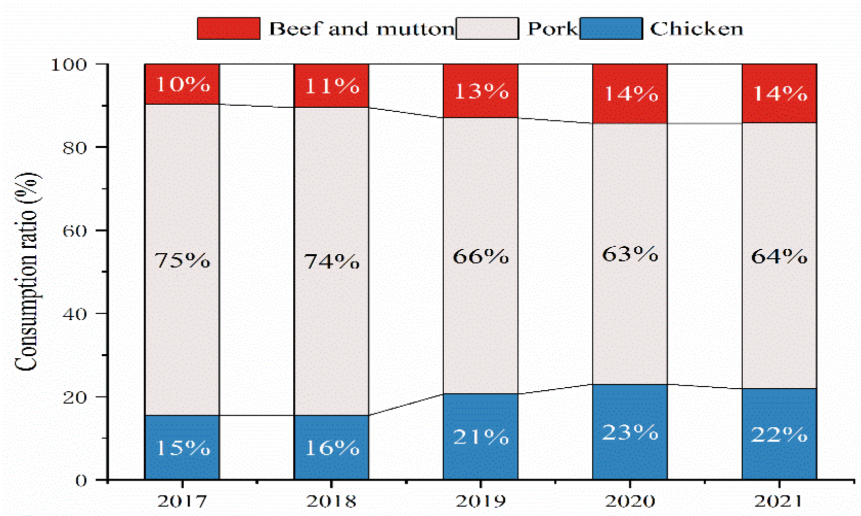

Figure 1.2 – Consumption structure of China's three major types of meat in 2017-2021

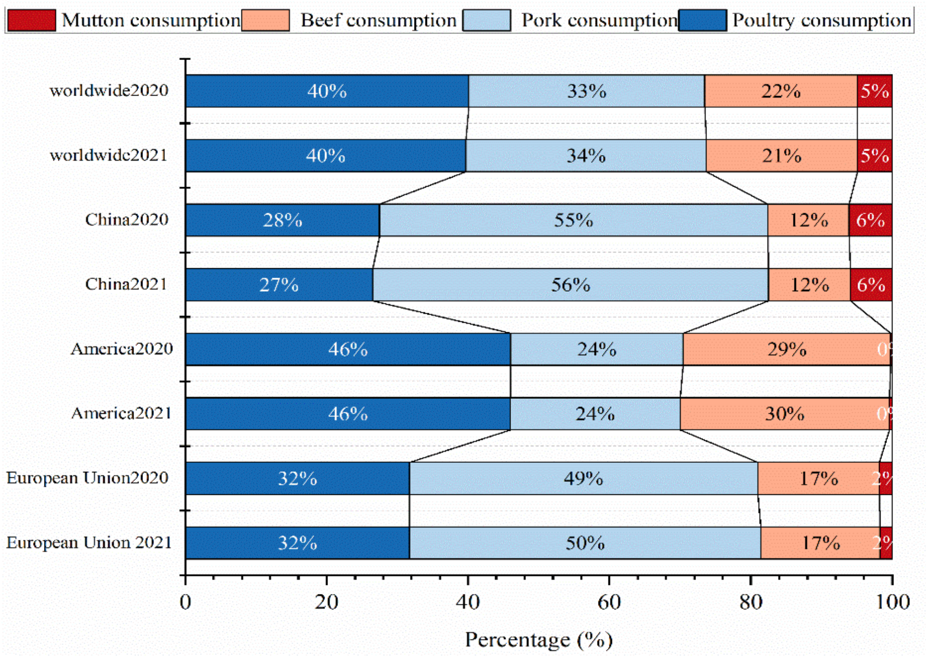

Figure 1.3 – Percentage of global meat consumption in 2020 and 2021

(2) Development trend of meat and meat products consumption

From the perspective of the world, meat products generally account for more than 45% of meat consumption, while in 2018, China's meat products consumption accounted for only about 20% of meat. Compared with the world level, China's meat products consumption still has more than twice the development space. There are two types of meat products: low-temperature meat products and high-temperature meat products. Low-temperature meat products are processed and preserved at lower temperatures, which can maximize the original flavor of meat products. This kind of meat product has the characteristics of fresh, tender, soft, delicious flavor, and its processing technology is advanced, which is superior to high-temperature meat products in quality. With the strengthening of consumers' concept of a healthy diet, low-temperature meat products will occupy a dominant position in the meat products market. In recent years, low-temperature meat products are gradually favored by more and more consumers, and have developed into a hot spot in the consumption of meat products. As shown in Figure 1.4, low-temperature meat products are also popular all over the world. Compared with high-temperature meat products, the consumption proportion of low-temperature meat products in Japan, the United States and the United Kingdom has reached more than 93%, while that in China is only 65%, which is far lower than those in developed countries.

Therefore, the proportion of low-temperature meat products in China will continue to increase.

Traditional meat products have high fat content, saturated fatty acid content, and cholesterol content, which increase the health problems of consumers. At the same time, they do not contain dietary fiber, which is detrimental to the emptying of food residues in the intestines, and contain preservatives, nitrites, etc., which restrict the development of meat industry to a certain extent.

Consumers in the 21st century have put forward high requirements for food and its health benefits. Therefore, developing new healthy meat products to meet people's growing demand for healthy food has become a new trend in the development of meat products, and low-fat meat products is one of the new trends.

Overall, it can be predicted that low-fat meat products will be a hot research direction in the development of low-temperature meat products in the future.

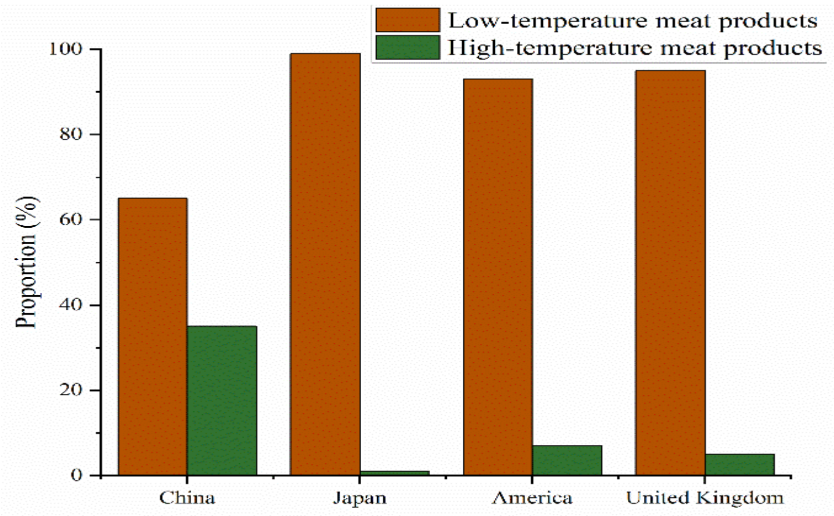

Figure 1.4 – Proportion of low-temperature and high-temperature meat products in China and major countries in the world in 2018

1.2 Analysis of the characteristics of low-temperature meat products and the progress of fat reduction technology

1.2.1 Low-temperature meat products

In China, there are four types of low-temperature meat products: meat filling products, western-style ham products, sauce-braised meat products, and roasted meat products. Among them, up to 20-30% fat is often incorporated to improve the emulsification effect in the production process of meat filling products. So emulsified meat products is the main field of research on low-fat meat products. Emulsified meat products include frankfurters, bologna sausages, Vienna sausages and other meat products, the most representative of which are frankfurters and bologna sausages. Emulsified meat products are also more and more popular among consumers in China due to their soft, juicy and attractive taste. It is one of the first-choice meat products both at home and in restaurants. As a result, emulsified meat products have become an integral part of our diets and have great economic benefits for meat processing companies as well.

In the production process of the emulsified meat products, lean meat with salt are chopped firstly to dissolve the myofibrillar protein. Then water and fat are incorporated, followed by continued chopped. According to the oil-in-water theory, the salt-dissolved myofibrillar protein can absorb a large amount of water and form an interface or film on the surface of the fat ball, that is, an interface protein film, which improves the emulsion stability of the fat. Meanwhile, some non-protein additives, such as starch or soy protein isolate, are also incorporated to the mixed system to improve the viscosity of the system and ensure the stability of moisture and fat in the system.

During the subsequent heating process, the high temperature induces the denaturation of myofibrillar proteins to form a three-dimensional network of gel in which water and fat are firmly bound.

At present, the mechanism of gel of emulsified meat products has been extensively studied, among which the most classic theories are the oil-in-water theory and the physical mosaic model [7, 8]. Both theories are intended to explain the stabilization mechanism of fat in the meat-emulsion system. According to the oil-in-water theory, fat is a dispersed phase, myofibrillar protein aqueous solution is a continuous phase, and myofibrillar protein acts as an interface protein-membrane to improve fat stability in the mixed system. According to the physical mosaic model, the size of fat particles in the meat batters system far exceeds the standard of the classical emulsification system (the diameter of the dispersed phase is less than 20 μ m), and fat particles are embedded in the sticky minced meat system by physical action. Some scholars also reported that the stability of fat particles in meat batters is the result of the joint action of interface protein-membrane and physical inlay fixation.

1.2.2 The role of fat in emulsified meat products

Fats composed of triglycerides are an important raw material for the processing of traditional emulsified meat products. Typically, ground meat products contain about 20-30% fat to obtain a good taste [9-11]. Fat affects the texture and flavor of meat products, as well as the gastro neurohormone pathway, which is in charge flavor perception and lubrication effect [12, 13].

Factors such as tenderness, juiciness, and flavor of emulsified meat products [14, 15] are the first factors consumers consider when choosing products. The addition of fat is beneficial to improve the flavor and texture of emulsified meat products[10].

The flavor is composed of the most basic flavors such as sour, sweet, bitter, spicy and fresh, as well as various volatile aromas. Different kinds of meat have their unique aroma. The taste and aroma of processed meat products develop primarily during processing. Lipids are the main source of flavor components in processed meat products, the half of the volatile flavor compounds in processed meat products forms directly or indirectly by lipids during the heating process.

The saturated fat of animal species can directly (degrade) or indirectly (reacted with proteins) to generate substances such as aldehydes, ketones, alcohols, free fatty acids and hydrocarbons [16] during the heating process to produce the characteristic taste of various meat. In addition, the detection threshold of these flavor substances is low, which makes an important contribution to the flavor of emulsified meat products.

The texture of emulsified meat products is a "key" feature that determines whether the product accepted by consumers. The texture features include the hardness of myofibrillar protein gel and the juiciness produced as fat particle exudation. This is a dynamic characteristic experience in the sensory evaluation process, including the shape, size and surface oiliness of emulsified meat products in the early chewing stage; tenderness and juiciness of the product during chewing; viscosity and swallowing sensation of product mixture after chewing[10].

In the process of chopping and mixing, the fat was dispersed into little fat balls, which are wrapped by salt-soluble protein (myofibrillar protein) and stabilized in the minced meat system. During the heating process, these fat balls were further bound by the three-dimensional network structure formed by the denaturation and cross-linking of myofibrillar proteins. Structurally, these protein-coated fat particles are filled in the protein gel network structure in the form of copolymers, reducing the voids in the meat product gel, and thus improving the gel strength [17, 18].

1.2.3 Harm of excessive intake of fat

In China, as people's living standards continue to improve, consumers' understanding of the relationship between diet and health continues to grow, and this increased focus on healthy food choices has led to changes in daily eating habits. Therefore, consumers' demand for healthy food is increasing day by day. Healthy food has strict control over some substances, such as fat, salt, caffeine or cholesterol. Although fat is one of the important nutrients that the human body needs to maintain daily life, up to 30% of animal fat improve flavor and texture of emulsified meat products, but animal fat is rich in saturated fatty acid and cholesterol. Excessive intake of saturated fatty acids and cholesterol will contribute to various health problems, such as hypertension, hyperlipidemia, cardiovascular diseases, obesity and some types of cancer [19]. The World Health Organization has also made recommendations for its daily intake. 1) Fat intake should not exceed 30% of total calorie intake; 2) Saturated fatty acids intake should not exceed 10% of the total energy intake; 3) The daily intake of cholesterol is less than 300g [20]. Because consumers gradually stay away from traditional emulsified meat products (high-fat meat products) to meet its green and healthy needs and gain market share, the meat processing industry has also re-optimized the formulation of emulsified meat products to reduce fat content or optimize fatty acid composition, such as replacing saturated fatty acids with unsaturated fatty acids.

1.2.4 Quality defects of low-fat meat products

As mentioned above, fat has an important influence on the sensory properties of emulsified meat products. On the one hand, the fat particles are filled in the protein gel network structure in the form of copolymers, which reduces the voids in the meat product gel, thereby improving the performance of the water holding capacity, mulsionsion stability, and gel strength of meat products, so that the emulsified meat products show good texture and juiciness. On the other hand, fat is the main source of flavor substances in emulsified meat products, and fat can directly or indirectly produce volatile flavor substances. Generally speaking, when it is observed that the flavor (taste and smell) decreases due to the reduction of any food component in the traditional meat product formula, the product will be rejected by consumers most of the time.

The appearance of meat products is one of the main factors to promote consumer choice. Fat and lean meat in meat patty can be identified by naked eyes. However, the structure of raw materials such as emulsified meat products is chopped fully, and it is difficult to distinguish the fat and lean meat in the chopped raw materials. The approximate fat and lean content can only be perceived through different color differences[21]. By increasing the moisture content in minced meat to make up for the reduced fat content, the appearance color of meat products will look darker. Because consumers usually associate dark color with products with high lean meat content, such appearance is beneficial to the sales of low-fat meat products, but at this time, the quality of emulsified meat products drops sharply.

In conclusion, simply reducing fat levels in emulsified meat products directly affects the emulsion stability, cooking yield, texture properties, and flavor of meat products. To achieve the product quality of traditional emulsified meat products, it is necessary to adjust or add fat substitutes to the various components in the low-fat emulsified meat products to maintain the original quality attributes, such as color, flavor, texture, and binding properties.

1.2.5 The main ways to improve the quality of low-fat meat products

To improve the quality of low-fat emulsified meat products and maintain customer acceptance, safety, texture and stability, meat process companies generally follow the following two basic methods. First, the easiest option is to replace part of the pork back fat with leaner meat, using a meat ingredient that ensures the proportion and functional properties of a low-fat emulsified meat product. This method means an increase in production costs, making this method unacceptable to most enterprises in the industry. Second, add little or no calorie water or other low-calorie non-meat fat substitutes[19]. The addition of non-meat materials is required to help improve product texture, especially some non-meat materials should have high water holding capacity, such as flour, starch, fibers [22], gums (xanthan, carrageenan, konjac) [23], proteins (e.g. hydrolyzed collagen, soy protein isolate)[9], other water-based ingredients including gels and hydrocolloids, and rich in polyunsaturated fatty acids lipid source [10]. This method of using fat substitutes can be assisted by novel processing techniques. For example, using ultra-high pressure techniques[24], chopping techniques[25], and ultrasonic techniques[26] to induce certain functional characteristics or alter the final product quality.

1.2.6 Current research on non-meat fat substitutes

Non-fat ingredients or healthy fatty acids (unsaturated fatty acids) as replace fat substitute are the most widely used approach in developing low-fat emulsified meat products. Current research on fat substitutes mainly focuses on three substances: carbohydrates, proteins and vegetable oils.

(1) Carbohydrate-Based fat substitutes

Carbohydrates are mainly polysaccharides and polysaccharide derivatives formed by glucose or galactose through 1, 3-glycosidic or 1, 4-glycosidic bonds. Carbohydrates have strong hydration capacity, and even some carbohydrates can form weak gel through hydration [27]. These fat substitutes stabilize water in protein gel system through their strong water holding capacity and exist in the protein network structure in the form of filled particles. At present, such fat substitutes mainly include gums (konjac gum[28], carrageenan [29], and xanthan gum[30]), dietary fibers (wheat dietary fiber [31], citrus dietary fiber [32], sugarcane dietary fiber [33-36], regenerated cellulose fiber [37], bamboo shoot dietary fiber [38], oat dietary fiber [36], pineapple dietary fiber [39], kiwi insoluble dietary fiber [40], bacterial nanocellulose [41]), starch (cassava starch, modified starch[10]) and other carbohydrates (inulin[42], microcrystalline cellulose[19]).

(2) Protein-based fat substitutes

As a fat substitute, protein may interact with the three main components (protein, fat and water) in low-fat emulsified meat products due to its lipophilic and hydrophilic properties. For example protein can interact with the polar groups of the original protein in meat or forming hydrogen bonds with free water, thus improving the gel properties and water holding capacity of low-fat emulsified meat products[11]. At present, collagen, egg white protein, chicken protein isolate, hydrolyzed collagen, concentrated milk protein, bovine plasma protein, as well as various plant proteins has been studied as fat substitutes [10, 19, 43]. Some plant proteins, such as legumes, cereal, oilseeds, mycoprotein, and soy protein groups (textured, flour, concentrate, and isolate) [44], have emulsifying, water / oil absorption capacity, and high nutritional value (such as balanced amino acids, essential fatty acids, vitamins and digestibility), making it a unique fat substitute [44, 45].

(3) Vegetable oil-based fat substitutes

When considering the texture and sensory characteristics of low-fat meat products, its overall nutritional status is also an aspect that can not be ignored. The overall nutritional status of meat products are affected by factors such as polyunsaturated fatty acid/monounsaturated fatty acid (PUFA/MUFA) content and its ratio, n-6/n-3 fatty acids and so on. Compared with animal fats, vegetable oils contain more PUFA and MUFA, some vegetable oils, such as canola oil [46], grape seed oil and corn oil [47], linseed oil [48], soybean oil [49], camellia oil [50], palm oil [51], peanut oil and safflower oil [52] ,fish oil [29], sunflower [53], olive oil [54] et al., have been reported as fat substitutes.

The addition of vegetable oils resulted in reduced oxidative stability of meat products due to the presence of unstable double and triple bonds in MUFA/PUFA. Pre-emulsification, encapsulation and the use of synthetic antioxidants are strategies to overcome lipid oxidation problems. Among them, pre-emulsification is the most commonly used method, which is a preparation process in which an emulsifier is used to stabilize the emulsion. In this case, the oil is dissolved in water (O/W), this process can improve the fat-binding capacity, enhance the physical stability, and make the pre-emulsified vegetable oil easier to disperse into the aqueous system. The emulsifier used is usually a protein of non-meat origin, such as soy isolate, sodium caseinate, and whey protein isolate. This pre-emulsified vegetable oil always prepared the day before the processing of the low-fat meat product and is incorporated to the meat product as a fat component. After pre-emulsification, vegetable oil forms smaller fat globules with increased specific surface area, resulting in more proteins are bound to the surface of the fat globules, thereby interacting with the meat matrix to form better bonds. This makes more protein in meat available for gel formation of the matrix, resulting in a stronger gel. In addition, structured oils have been a research hotspot in recent years. For example, oleogels are an interesting option to incorporate vegetable oil (fat substitute) into meat emulsions to develop healthy meat products with good physicochemical properties. Oleogels are characterized as semi-solid systems with a liquid hydrophobic phase immobilized into a three-dimensional network of lipophilic solids (gelators). Monoglycerides are economically viable among the existing gels, and the high oil content of sunflower, olive, soybean, linseed, and rapeseed oils have been successfully used to produce oleogels as a substitute for animal fat [55].

1.3 Analysis of production status of cultivated mushrooms and its application in food processing

1.3.1 Current market and commercial aspects of cultivated mushroom

Mushrooms are very popular and valuable food items in modern dietary regimes because of their nutritional value. Cultivated mushrooms have a balanced composition of nutrients, they also grow quickly, without requiring expensive equipment for growing, are easily processed, and have a pleasant taste and aroma in ready product.

Edible mushrooms are widely consumed in many countries as a food having a high commercial and culinary value, mainly due to organoleptic properties, such as texture and flavor, and it is possible to distinguish a mushroom species based on its characteristic odor or flavor [56]. Nowadays, the great interest in cultivated mushrooms commercialization is due to its flavor, taste, and several medicinal applications. Mushrooms represented a market of 63 billion US dollars[57]. Edible, medicinal, and wild mushrooms are the three major components of the global mushroom industry. Cultivated mushrooms are the most representative mushroom species at the market as shown at the Figure1.5.

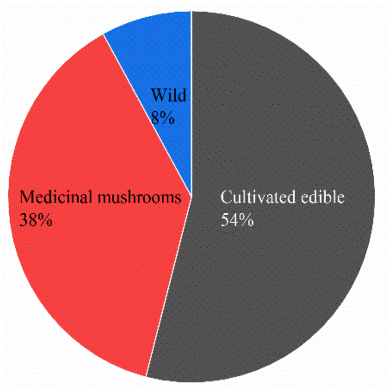

Figure1-5 – Market represents of edible mushrooms

According to Food and Agriculture Organization, mushroom cultivation worldwide in 2018 was approximately 9 million tons[58]. China edible mushroom annual production are shown at the Figure 1-6[59].

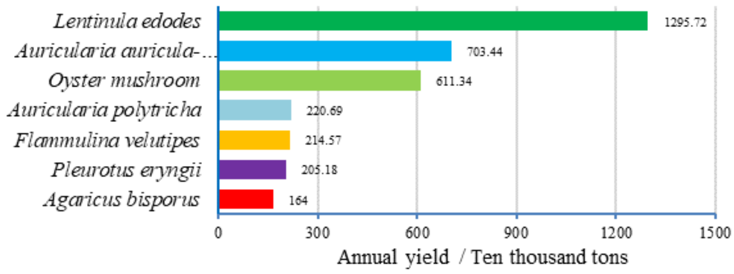

Figure 1.6 – China edible mushroom annual production by genus in 2021[59]

As shown in Figure 1.6, China mushroom varieties most commonly grown on a commercial scale with an annual yield of more than 1 million tons are *Lentinula edodes, Auricularia auricula-judae, Oyster mushroom, Auricularia polytricha*, *Flammulina velutipes, Pleurotus eryngii, and Agaricus bisporus,* with a yield of 1295.72, 703.44, 611.34, 220.69, 214.57, 205.18, and 164 ten thousand tons, respectively. Approximately 90% of the companies producing edible and medicinal mushrooms operate in China [56]. The nutritional and therapeutic use of mushrooms in Asian countries results from centuries of tradition and is much better documented than in Europe. Cultivation of edible mushrooms combines the production of protein-rich food with lignocellulosic organic waste recycling, thus meeting the pro-ecological strategies of modern agriculture[59].

Mushrooms are environmentally friendly. They biosynthesize their own food from agricultural crop residues, which would otherwise cause health hazards. Useful macrofungi consist of those with edible and medicinal properties. There is no easy distinction between the two categories. Many of the common edible species have therapeutic properties; several medicinal mushrooms are also edible [59]. A total of 126 medicinal functions are thought to be produced by medicinal mushrooms and fungi, including antitumor, immunomodulating, antioxidant, radical scavenging, cardiovascular, anti-hypercholesterolemia, antiviral, antibacterial, anti-parasitic, antifungal, detoxification, hepatoprotective, and antidiabetic effects [60].

1.3.2 Current application of cultivated mushrooms in food processing

Mushrooms are delicious and contain many flavor substances[61]， and are rich in proteins, amino acids, polysaccharides, dietary fiber, unsaturated fatty acids, nucleosides, mineral elements, and a variety of physiologically active substances, such as antioxidants ergothioneine, glutathione and β-glucan [62-66].

Mushrooms have important antioxidant effects [65, 67, 68]. The water extract and alcohol extract of *Coprinus comatus* have certain clear ability to DPPH radical, superoxide anion, hydroxyl radical [69] and also have hypoglycemic[70], antitumor, and immunomodulatory effects [71]， protective effects on the cardiovascular system [72], antibacterial, anti-inflammatory, weight loss and other medical care functions [63, 73-75].

Nowadays, people's pursuit of nutrition and health food is growing stronger, so mushrooms are favored due to their rich nutrition and health functions. Much research has focused on the development of mushroom products. Nowadays mushrooms are widely used in food industry, and consumers can purchase their favorite mushroom products from the market [63].

(1) Simple processed products

Simple processing is currently the main form of mushroom processing. Mushrooms have the high moisture content and short storage period, which are often processed into dry products [76, 77] and canned, pickled, and sugar products. These simple processing methods can better maintain the original shape and flavor of the mushrooms and increase the visibility of the product. The simple form of processing makes mushrooms safer, convenient and hygienic to eat [78].

(2) Nutraceuticals

Functional ingredients such as protein, polysaccharides, polypeptide, amino acids, and triterpenes in mushrooms can be used to produce into nutraceuticals. There are many studies on the extraction, structure and functional characteristics of mushroom polysaccharides, peptides, and protein, and some of them have been commercialized as health products [79-81].

Mushrooms are rich in polysaccharides[82], which have the function of anti-tumor and enhancing immunity. Nowadays, the biological effect of immune polysaccharides in mushrooms are getting more and more attention [79]. Such as *Flammulina velutipes* polysaccharide nanoparticles [83], *Pleurotus ostreatus* glucan [84], water-soluble polysaccharides, HPA and HPB from *Hericium erinaceus* [85], water-soluble polysaccharides from *Termitomyces eurhizus* [86]. Water-soluble polysaccharides(α-d-glucans) from the fruit bodies of *Coprinus comatus* [87].

Mushroom extract has antioxidant and some other special functions, which are often processed into nutraceuticals. For example, alcohol extract of *Pleurotus ostreatoroseus* [62]. β-glucan extracted from *Lentinus edodes* [88].

The abundant protein in mushrooms has good antioxidant, antitumor and immunomodulatory activities and can be processed into nutraceuticals. For example, the protein in *Pleurotus eryngii* can be extracted by enzymatic method to produce functional protein powder [89].

(3) Meat products

Meat products are one of the most common foods in people's daily life. However, long-term consumption of meat and meat products will cause health risks due to its high fat content. More and more people are inclined to low-fat meat products or vegetarian [90]. The high protein content and low fat content of mushrooms make them a cost-effective alternative to fat and meat in meat products [45] . For example, using *oyster* mushroom and glutinous rice to replace the pork of traditional Thai fermented sausage [91]. 77% sea cork fish and 20% *oyster* mushroom flour can make high-protein sausages with excellent color and flavor [92]. 3% brewer’s spent grain and 8% mushrooms can increase the protein content of smoked sausage and reduce the content of microorganisms [93]. Partial replacement of meat in sausages with boiled mushrooms or fried mushroom powder can reduce the amount of soy protein in sausages and the calories in sausages [94].

In addition, mushrooms are incorporated to meat products to produce functional meat products because of its rich functional components. For example, *Straw* mushroom can improve the physical properties of meat sausage, reduce its peroxide value, increase the content of protein and essential amino acids, and the ratio of polyunsaturated fatty acids to saturated fatty acids in sausage [95]. *Shiitake* added to the sausage as a substitute for some lean meat can increase the moisture, total dietary fiber, methionine, glutamic acid, cysteine, total phenol content, and the antioxidant activity of sausage [96]. Adding *shiitake* mushroom powder to low-salt chicken products can inhibit the oxidation of fat [97]. Adding *sun* mushroom powder can increase the oxidative stability of pork sausages [98], 2.0% *sun* mushroom ethanol extract significantly improved the oxidative stability of pork sausages, which can be stored at 4 ° C for 21 days [99].

(4) Beverage

Mushroom beverage refers to a type of products obtained by leaching, fermentation or direct processing of mushroom fruiting bodies, mycelia and their culture fluids, which have the nutritional value, flavor and health care functions of mushrooms [100]. Mushrooms can be made into a variety of beverages, such as mushroom fruit body formulated drinks, mushrooms and fruit juice, milk compound beverages, mycelia mixed drinks [101]. In addition, fermentation of mushrooms in the matrix can increase many metabolically active substances, making them richer nutrition and unique health functions. Therefore, mushrooms can also be fermented by adding strains to obtain mushroom fermented tea drinks, vinegar drinks, wine and other products[102].

(5) Seasoning

Mushrooms are rich in amino acids, and various amino acids will form unique flavor polypeptides [64]. Different mushrooms contain different polypeptides. The water extract of fresh Ab contained polypeptides Gly-Leu-Pro-ASP and Gly-His-Gly-ASP, which can improve the complexity, taste and palatability of chicken soup. Therefore, It is feasible to manufacture the thick peptide with Ab [103].Using mushrooms or scraps from the mushroom processing as raw materials, the mushroom cell wall is rapidly decomposed and broken by the enzymatic method, ultrafine crushing, ultrasonic extraction and other technologies[104], and the functional and umami substances are sufficiently released. By spray drying or fermentation with other auxiliary materials, various mushroom seasonings can be prepared, which are rich in amino acids and nucleotides, and have a strong fresh flavor, rich nutrition, and safety to eat. The more mature products are convenient soups, mushroom vinegar, and mushroom sauce [105]. In addition, the concentrated solution of mushroom enzymatic solution and animal protease made from fresh bone were mixed and prepared in a certain proportion, and the mushroom compound meat flavoring in the states of juice, sauce and powder could be manufactured through the Millard reaction [106].

(6) Other foods

Mushrooms can also be used to produce flavored snacks, or be added to other foods as auxiliary materials to increase the flavor and nutritional value, such as mushroom chips [107]，mushroom sauce [108], mushroom fermented milk [109], mushroom biscuits [110, 111], mushroom flavored snacks [112]，mushroom cake and bread [113], mushroom noodles [114, 115], mushroom semi-finished food [116, 117]. The above edible mushroom products are diverse and rich in nutrition, which has expanded the space for edible mushroom applications.

1.4 Analysis of health beneficial qualities and application prospects of Ab in low-fat meat products

1.4.1 The characteristics of Ab

Ab or button mushroom is the most widely cultivated and consumed mushroom throughout the world, contributing to approximately 40% of world mushroom production [76], and China is the country with the largest production of Ab mushroom in the world [57], which are often considered to provide a substitute for meat with a comparable nutritional value to many vegetables [118]. This is an edible basidiomycete mushroom. The nutritional value of the mushroom is one of the main factors determining its quality. Protein is an important constituent of mushrooms dry matter, carbohydrate represent the bulk of their fruiting bodies and their fat content is very low [119]. Given the crucial role that minerals play in the biological process and metabolism and their therapeutic use in dietary supplements, foods are often tested for nutrient minerals in relation to specific health benefits. So, Ab mushroom has an especially high content of phosphorus, sodium and potassium followed by Ca, Mg, Na, Fe and Zn. It is also an excellent source of several essential amino acids and vitamins (B2, niacin and folate)[56]. Nutritional composition, microelements and vitamins present of Ab mushroom [120] is given at the Figure 1.7, Figure 1.8, Figure 1.9.

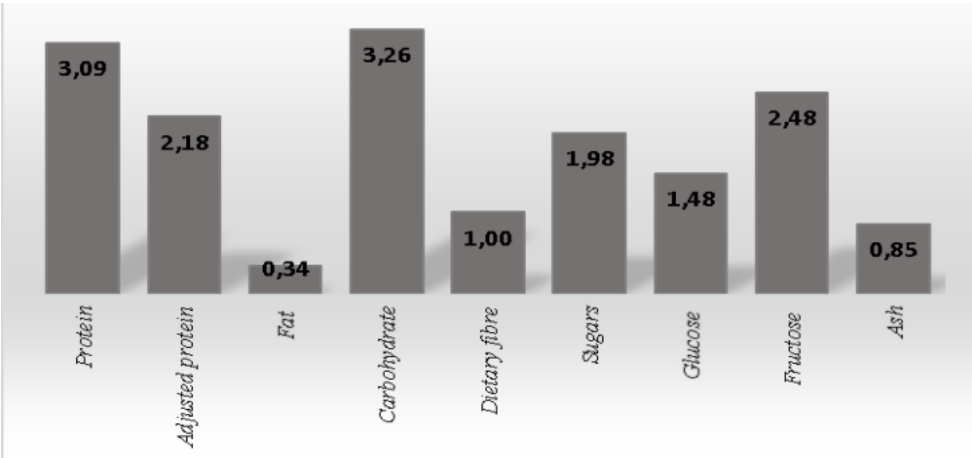

Figure 1.7 – Nutritional composition of Ab *mushroom* (unit/100 g)

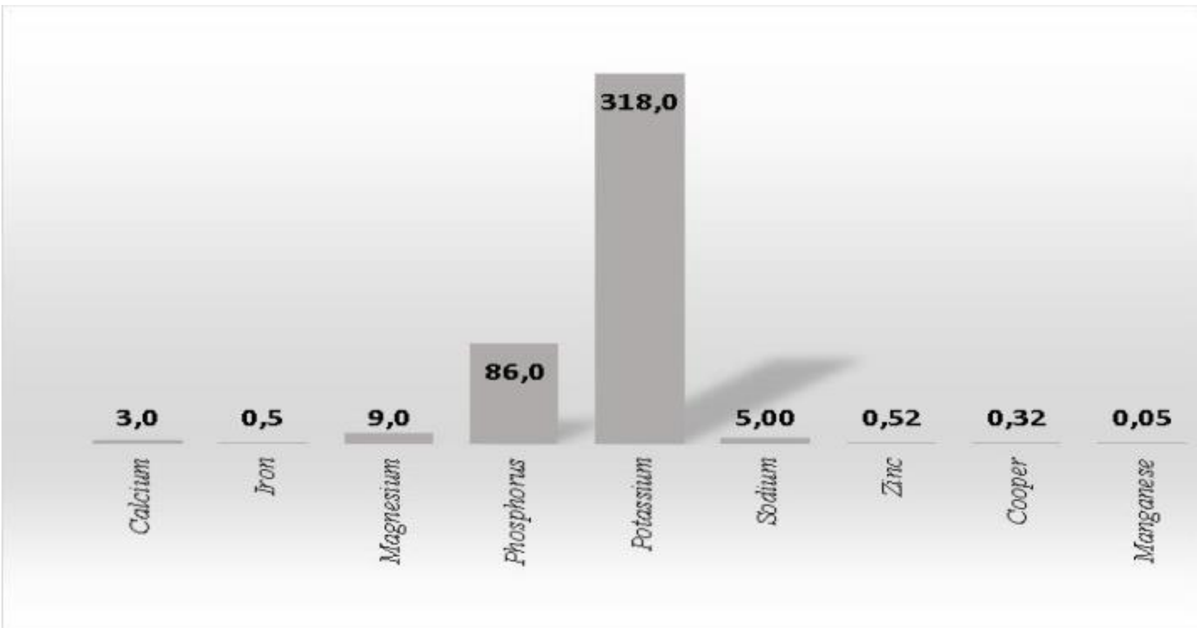

Figure 1.8 – Microelements present of Ab mushroom (mg/100 g)

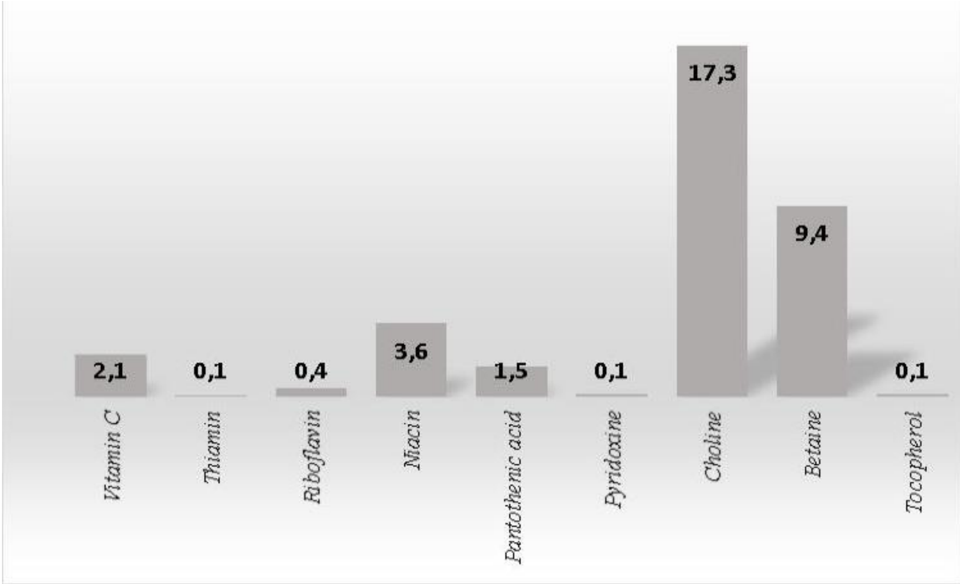

Figure 1.9 – Vitamins present of Ab mushroom (mg/100 g)

As can be seen from Figure 1.7, 1.8, and 1.9 that Ab mushroom has a rich nutritional composition, microelements and vitamins content. Besides them, around half of the fungal cell wall mass is constituted by β-glucans along with ergosterol, tocopherols, linoleic acid, and lectins. Also, they contain 1-6 mg of phenolics / g of dried mushroom and flavonoid concentrations ranged between 0.9 and 3.0 mg/g of dried matter; as myricetin and catechin [121].

Amino acids contents in Ab mushroom are vary and depend on the stage of development the mushroom. An amino acids content compared with the "Perfect protein" and “Egg protein pattern” is presented in Table 1.2 [122].

Table 1.2 – Amino acids content in proteins of mushroom, compared with the “Perfect protein” and “Egg protein pattern”

Amino acids	The content of this amino acid, g per 100 g of protein				
	Perfect protein	*Pleurotus ostreatus*	*Lentinula edodes*	*Agaricus bisporus*	Egg protein pattern
Total essential amino acid content	36.0	42.7	45.7	69.3	51.3
Valine	5.0	5.0	5.2	8.1	7.3
Isoleucine	4.0	3.8	4.7	16.4	6.6
Luecine	7.0	8.8	9.2	14.3	8.8
Lysine	5.5	5.0	5.6	13.0	6.4

Methionine +Cystine	3.5	7.5	6.0	4.3	5.5
Threonine	4.0	4.2	4.8	4.3	5.1
Phenylalanine +tyrosine	6.0	6.9	9.0	5.6	10.0
Tryptophan	1.0	1.5	1.2	3.2	1.6
Total nonessential amino acid content	-	54.0	55.2	78.0	
Arginine		5.4	5.3	8.8	
Glutamic acid		16.6	11.5	12.7	
Alanine		8.2	9.9	9.2	
Glycine		6.1	10.3	6.7	
Asparaginic acid		9.8	7.8	22.9	
Proline		1.7	3.4	8.2	
Serine		4.2	4.7	7.4	
Histidine		1.9	2.2	2.0	
Total amino acid	-	96.7	100.9	147.3	

As can be seen from Table 1.2, cultivated Ab mushroom have a significant content of essential amino acids and total amino acid, which is much higher than that of perfect protein and chicken protein, as well as *Lentinus edodes* and *Pleurotus ostreatus*. Its amino acid pattern is close to perfect protein. This allows us to predict the prospects of using this type of raw material as a source of high-grade protein. Due to their high nutritional value, Ab mushrooms has a significant therapeutic effect. This allows them to be classified as nutraceuticals[123]. They show antioxidant and immunomodulating activity, metabolic effect, anticancer activity, anticholesterolemic and antiglycemic effect, anti-inflammatory activity [124]. Also, they enhance the maturation of bone marrow derived dendritic cells. The powder of Ab have a beneficial effect on the intestine, antinociceptive properties, antimicrobial activity[118, 121, 125].

1.4.2 Application prospects of Ab mushroom in low-fat meat products

The consumption of meat products in the United States and China in 2018 is shown in Table 1.3 [126]. It can be seen from the Table 1.3 that meat products in the United States are dominated by bacon (33%) and sausage (28%), followed by canned pork (16%) and ham (15%). Meat consumption in China is dominated by China-specific ham (29%), followed by ham (20%) and canned pork (21%) and sausages (14%). Therefore, ham, sausage, and canned food are very popular in China and the United States. Since only sausages are produced with fat added separately, considering the feasibility of processing, sausage is more suitable for low-fat meat products. Sausages are low-temperature meat products. As the result of the previous analysis, consumers' demand for low-temperature meat products is gradually increasing. Therefore, sausage will have a lot of room for development in the future. As previously analyzed, chicken products are becoming more and more popular. It is foreseeable that the demand for chicken sausage will gradually increase. However, in the production process of chicken sausage, high content of animal fat is often added in order to maintain emulsion stability, reduce cooking loss, improve water holding capacity, provide juice, hardness and flavor, which affects consumers' perception of the product to a certain extent. As mentioned above, Ab is delicious, nutritious, rich in protein, polysaccharides, dietary fiber, trace elements and essential amino acids, making it an ideal choice for fat replacement.

The hypothesis that Ab mushroom powder can used as a fat substitute to improve the quality of emulsified chicken products is based on the fact that Ab mushroom is rich in the following components:

(1) Protein, hydrophilic and hydrophobic side chains in the protein structure is responsible for surface activity, making Ab mushroom hydrophilic and lipophilic and forming gels.

(2) Polysaccharide, which forms an extended network in the continuous phase by increasing the viscosity and acts as an emulsion stabilizer.

(3) Fiber, the water-binding properties of the fiber make it possible to reduce cooking losses and improve water holding capacity. Meanwhile, dietary fiber, as an effective filler to improve the strength of protein-protein matrix, can enhance the gel properties of protein.

(4) The unique flavor of Ab mushroom can make up for the loss flavor of sausage caused by fat reduction. At the same time, except for producing flavor substances during the production of Ab powder, the color of Ab powder will deepen due to browning, and this unique color can reduce the use of nitrite. The umami of Ab mushroom also adds flavor to the sausage, thereby reducing the amount of salt. The low-cholesterol and low-

fat characteristics of Ab mushroom also reduce the cholesterol content and calories in sausages.

In conclusion, Ab mushroom is an ideal fat substitute, which can meet consumers' demand for healthier meat products with low fat, low salt, low cholesterol, low nitrate and low calorie. Therefore, Ab mushroom will have a very broad application prospect in low-fat chicken sausage.

Table 1.3 – Percentage of meat products consumption in the United States and China in 2018 [126]

Meat products	United States	China
Ham	15%	20%
Bacon	33%	3%
Sausage	28%	14%
Canned pork	16%	21%
Other	8%	13%
Ham sausage		29%

CONCLUSIONS OF SECTION 1

1. Meat is rich in protein, fat and microelement. Among the common types of meat, the fat content of chicken is relatively low. From the perspective of nutrition, chicken should be consumed more, especially chicken breast. Chicken consumption will continue to grow. Low-temperature meat products dominate in the consumption structure of meat products and will be the future consumption trend and research hotspot.

2. Low-temperature meat products can maximize the retention of nutrients in meat. Fat plays a key role in the quality of low-temperature meat products, but excessive intake of fat will be detrimental to health. Directly reducing the fat in meat products can lead to a decrease in the quality of meat products. Using non-meat fat substitutes such as protein, carbohydrates, vegetable oils, etc. can solve this problem.

3. Top five cultivated mushroom production in the world are *Lentinula, Pleurotus, Auricularia, Agaricus,* and *Flammulina gener*. Mushrooms are high-protein and low-fat foods, and rich in functional ingredients, which are widely utilized in food. To the knowledge of the author, there is no research report on Ab mushroom as a fat substitute.

4. Ab mushroom was rich in protein, its amino acid content is close to ideal protein, and it is rich in carbohydrates, potassium, phosphorus and other microelement, as well as

health ingredients such as choline and betaine, yet low in fat. The unique nutritional properties of Ab mushroom make it ideal for the production of low-fat healthy meat products. In view of the continuous growth in the consumption of chicken and low-temperature meat products, Ab mushroom will have a broad application prospect in the production of low-fat chicken sausage in the future.

SECTION 2 MATERIALS AND METHODS OF THE RESEARCH

2. 1 Experimental materials

Fresh Ab mushrooms, chilled chicken breast and pork back fat were purchased from Hualian Supermarket of Xinxiang City. Salt, sodium tripolyphosphate, white pepper, sugar, soybean oil, chicken essence, potato starch, 25 mm collagen casings and ice water were all food grade. All other chemicals, including Tris, EDTA, KCl, $MgCl_2$, NaCl, K_2HPO_4, KH_2PO_4, EGTA, NaN_3, and KBr were analytically pure compounds.

2. 2 Research methods

2.2.1 Preparation of Ab powder

Ab were washed using tap water, drained the water on the surface, cut into 3-5 mm thick slices, then put into the oven and dried at 45 °C for 8 hours until the moisture content was 7%. After natural cooling, the dried Ab mushrooms were crushed with a high-speed grinder. Then the mushrooms powder was sieved through a sieve of 80 mesh, 120 mesh, 160 mesh, and 200 mesh sieves in sequence. The corresponding screen underflow was obtained, which was packed using a PE plastic bag and placed in a desiccator for subsequent experiments.

2.2.2 Pretreatment of chicken breast meat and pork back fat

All visible connective tissue and fat were trimmed from the chicken breast meat. Furthermore, all visible connective tissue and the remaining lean meat were cut from pork back fat. The chicken breast meat and pork back fat were ground separately using a grinder (MM-12, Guangdong, China) with a 6 mm holes plate. Every 500 g of minced meat and 200 g of pork back fat were packed in nylon/polyethylene bags, sealed, stored (−20°C), and used up within 4 weeks.

2.2.3 Preparation of chicken batters

The thawing chicken breasts (0−4°C) was chopped with salt, tripolyphosphate, and 1/3 ice water by a chopper at 1500 rpm for 60 seconds. After a 3 minutes pause, ground chicken was chopped with thawing pork back fat, Ab, sugar, ground white pepper, and 1/3 ice water depending on the formula by a chopper at 1500 rpm for 120s. After another 3 minutes pause, ground chicken was chopped with the remaining 1/3 ice water at 3000 rpm for 60 seconds. During the chopping process, it was necessary that the center temperature of the mixture was always lower than 10°C. The process flow chart of chicken batters production is shown in Figure 2.1.

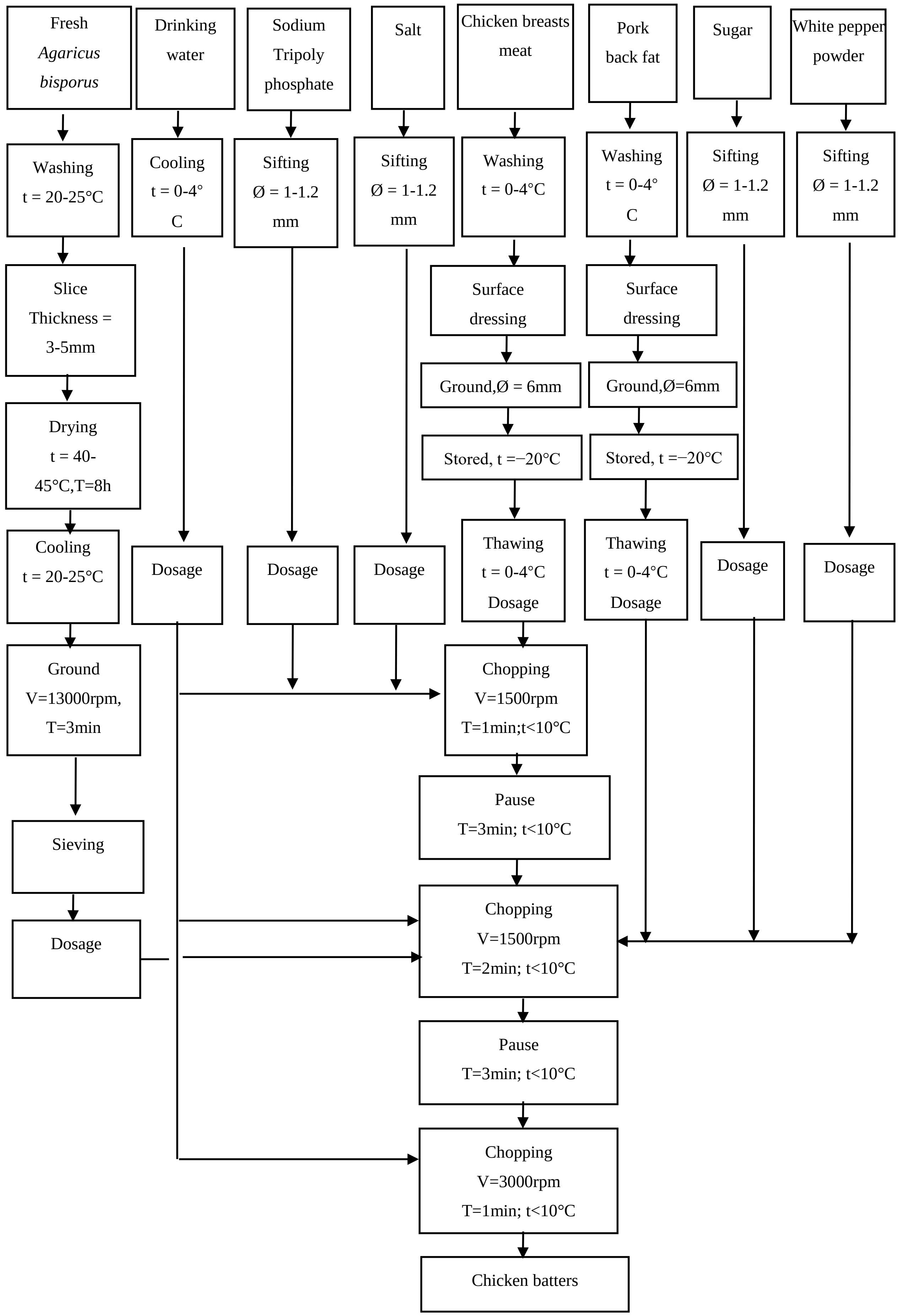

Figure 2.1 – Process flow chart of chicken batters production

2.2.4 Extraction of myofibrillar protein (MP)

The MP was extracted from raw breast chicken by sodium phosphate buffer (20 mmol/L K_2HPO_4/KH_2PO_4, 0.1 mmol/L NaCl, 2mmol/L $MgCl_2$, 1 mmol/L EGTA, 1 mmol/L NaN_3, 100 mmol/L KCl, pH 7.0, 4 °C) according to the method of [127]. The protein content was determined by the biuret method [128]. Bovine serum albumin (BSA) was used as standard substance.

2.2.5 Manufacture of low-fat sausages with Ab

Ab powder was mixed with soybean oil at a ratio of 1:2 for 24 hours to absorb oil and expand. The preparation method of chicken batters as described in 2.2.3. Then, chicken batters were stuffed into 18 mm collagen casings forming 10 cm frankfurters. The uncooked sausages were baked at 60°C for 30min and cooked them in a temperature-controlled water at 80 °C for 30 min. Next, the cooked sausages were vacuum-packed (QS-DZ500, Shandong Quansheng Machinery Co., Ltd., China) in each polyethene bag and heated in an 80°C water bath for 15 seconds. Finally, the sausages were cooled in an ice water bath and stored at 4 °C until the laboratory analysis.

2.2.6 pH determination

The pH value was determined carried out as described by Choe J et al. (2018) [129] with some modification. 10 g of sample were mixed with 100 g of 0.1mol/L potassium chloride solution, the mixture was homogenized (speed, 8000 rpm) for 1 minute in homogenizer (T25, IKA, Germany). The homogenates were filtered through Whatman No. 4 filter paper (Whatman, Maidstone, England), the pH value of the filtrate was measured with pH meter (Model320, Metler-Toledo Ltd, Essex, UK).

2.2.7 Emulsion stability determination

According to the method of Fernandz Martin et al. (2009) [130] , 25g of chicken batters were loaded into a 50 ml centrifuge tube, and centrifuged at 500xg for 5 min at 3°C to remove air bubbles in the meat batters. The closed centrifuge tubes were heated in a water bath at 80°C for 20 min. Next, remove the tube, open the lid, and place the tube upside down on a plates for 50 minutes at room temperature to release any exudate. The exudate was called cooking loss and expressed as % of the initial sample weight. The water loss was expressed as % of the initial sample weight and determined from the dry matter content of total exudate after heating at 105 °C for 16 h. The fat loss was calculate as the difference between cooking loss and water loss, which ignored any minor protein or salt components. Each treatment group was repeated 4 times.

2.2.8 Texture analysis (TPA)

40 g of chicken batters were loaded in a 50 ml capped plastic centrifuge tube and heated at 80°C for 30 min in a water bath (the core temperature was 72°C) after centrifugation at 500×g for 5 min (Sorvall LYNX4000, Thermo Fisher Scientific, Germany). After that, the tubes were cooled to room temperature and then kept overnight at 2 ± 2 °C until testing. For chicken sausage, TPA proceeded directly after room temperature was restored. The samples were cut into the cylindrical-shaped (diameter, 25 mm; height, 20 mm) after returning to room temperature. The texture profile analysis was carried out at room temperature in five replicates for each formulation by the TA-XT plus texture analyzer with a probe P/36R (Stable Micro Systems, London, UK) , equipped with a 1 kg load cell, and the calibration distance was 30 mm. The settings used for texture analysis were as follows: pre-test speed, 2mm/s; test speed, 2.0mm/s; post-test speed, 5mm/s; trigger type, auto-5 g; time, 5s; and strain, 50%. The hardness, springiness, cohesiveness and chewiness were analyzed.

2.2.9 Color determination

The color measurement was carried out in five replicates for each formulation using a Minolta chromameter (CR-400, Minolta Camera Co., Japan). The standard white colorimetric plate was L* = 93.56, a* = 0.01, b* = 3.45. The probe of the chromameter was placed close to the surface of the chicken gel (height = 20 mm, diameter = 25 mm) to avoid light leakage. The Hunter's color values [L^* (lightness), a^* (redness), and b^* (yellowness)]of three chicken gel from each treatment were determined, and the averages were reported. The whiteness value was calculated according to the previously reported method [131].

$$Whiteness = 100 - \left[(100 - L^*)^2 + a^{*2} + b^{*2}\right]^{\frac{1}{2}} \quad (1)$$

2.2.10 Microstructure observation

The microstructure was observed using a field emission scanning electron microscopy (SEM) (Quanta 200, FEI CO., US) according to the method proposed by Wattanachant et al. (2005) [132] with some modification. First, the chicken gels (3 ×3 × 3 mm^3) were fixed with glutaraldehyde (2.5%) in phosphate buffer saline (pH 6.8) at 4 °C for 24 h. The fixed samples were washed thrice with 0.1 M phosphate buffer (pH 6.8) for 15 min. The washed samples were dehydrated sequentially in 50%, 60%, 70%, 80%, and 90% ethanol for 15 min and then in 100% ethanol for 10 min (thrice). The dried samples were then degreased with chloroform for 1 h. The degreased samples were replaced with a mixture of absolute ethanol and tert-butyl alcohol (volume ratio =1:1) and then with tert-butyl alcohol for 15 min. Next, the samples were vacuum dried at 45 °C for 5 h. Finally, the vacuum-dried sample was mounted on a bronze stub and sprayed with gold particles. The gold-plated samples were imaged under a microscope at 5 kV. Many micrographs were taken to select the most representative ones.

2.2.11 Determination of cooking yield (CY)

The exudate separated from the chicken gel was removed after storage at 4°C overnight. Next, the cooked batters were weighed, and CY was estimated as % weight yield during cooking, expressed as the ratio of cooked batters (g) to raw batter weight (g).

2.2.12 Determination of water holding capacity (WHC)

W_1 grams of chicken gel were wrapped in filter paper and placed in a 50 mL polypropylene tube. The chicken gel was then centrifuged at 8000 rpm for 10 min at 4 °C. Next, the sample was removed from the centrifuge tube, the filter paper was carefully removed, and the weight of the chicken gel after centrifugation (W_2) was obtained. WHC (%) was the percentage of the gel weight after centrifugation (W_2) relative to the original gel weight (W_1).

2.2.13 Determination of rheological properties

The rheological characteristics of chicken batters were measured using a rheometer (HAAKE MARS III, Thermo Scientific, Massachusetts, USA) according to a previously reported method [25] with some modifications. The chicken batters were placed between two parallel plates (diameter = 40 mm; gap = 1 mm), and the edge of the sample was sealed with methyl silicone oil to avoid contact with air. A temperature ramp sweep was conducted by heating the sample from 20 °C to 80 °C at the rate of 2 °C/min after holding for 2 min. The batters were continuously sheared in an oscillatory mode at a shear frequency of 0.1 Hz. The storage modulus (G') at different temperatures was recorded to characterise the rheological properties of the chicken batters. Each treatment was tested three times.

2.2.14 Determination of low-field nuclear magnetic resonance (LF-NMR)

For each treatment, approximately 3 g chicken gel was placed in a 25 mm diameter NMR probe to measure the spin-spin relaxation time (T_2) using an NMI20-040V-I Series NMR analyzer (Suzhou Niumag Co., LTD, Suzhou, China). During the data collection, the transverse relaxation curve was collected with CARR Purcell Meiboom Gill (CPMG) pulse sequence. The test parameters were set as follows: pulse interval, 0.5 ms; main frequency, 20 MHz; offset frequency 625998.37 Hz; TD,600008; NS, 8; TW,5500 ms; τ = 6.52 μs; and temperature, 32 °C. The attenuation curve of CPMG was inversed by T_2 inversion fitting software, and the relaxation spectrum and T_2 were acquired. To increase the reliability of measures, each treatment was tested three times.

2.2.15 Determination of gel strength

Gel strength analysis was performed using a texture analyzer (TA-XT Plus, Stable Micro Systems Co. Ltd., London, UK). The gels underwent a compression test using a cylindrical aluminium probe (P/0.5 inch). The settings were as follows: pre-test speed, 2 mm/s; test speed, 1.0 mm/s; post-test speed, 1 mm/s; interval time, 5 s; distance, 4 mm; trigger force, 5 g. Each treatment was tested five times. Each treatment was tested five times.

2.2.16 Secondary structure analysis of MP

Ab-MP gels were dehydrated completely in a lyophilizer (Alpha -2LD PLUS, Christ, Germany). Next, the lyophilized sample was ground into a fine powder, fully grounding with KBr, and made into thin sheets under high pressure for scanning. FTIR spectra were obtained using a spectrophotometer (Tensor 27, Bruker Optics, Inc., Karlsruhe,Germany) according to the method of [133]. All spectra were recorded at ambient temperature and scanned for sixteen times. PeakFit software (version 4.12, Systat

Software Inc., San Jose, CA, USA) and Origin software (version 8.6, Origin Lab, Chicago, IL, USA) were used to process these spectra. The percentages of the four conformations of α-helix, β-sheet, β-turn, and random coil were obtained, which represented the secondary structure of the protein.

2.2.17 Sensory evaluation

The sausages were subjected to sensory evaluation after being stored in a refrigerator at 4°C overnight. Sensory properties were evaluated by 50 males and 50 females. Sausages were cut into small cubes of the same size. Then, sausages were placed on a white plate. The temperature of the samples returned to room temperature before sensory evaluation. Each sample was numbered with random 3 digits (eg: 376; 258; 429, etc.). A 9-point happiness scale was employed to evaluated the samples (extremely like = 9; very like = 8; somewhat like = 7; somewhat like = 6; neither like nor dislike = 5; somewhat dislike = 4; moderately dislike = 3; dislike very much = 2; dislike extremely = 1). The sausage was scored for color, flavor, taste, texture, and general acceptance.

2.2.18 Proximate composition determination

The content of moisture, ash, protein and lipid were determined according to the Association of Official Analytical Chemists[134]. All determinations were performed in triplicate using three samples for each treatment.

The total carbohydrate were calculated from the content of water, protein, lipid and ash according to the following formula [93].

$$\text{Total carbohydrates (g/100g)} = 100 - (\text{g moisture} + \text{g protein} + \text{g lipid} + \text{g ash}) \quad (2)$$

2.2.19 Energy value determination

The energy value was calculated from the content of protein, carbohydrates and lipid according to the following formula[93].

$$\text{Energy value (kcal/g)} = 4 \times (\text{g protein} + \text{g carbohydrates}) + 9 \times \text{g lipid} \quad (3)$$

2.2.20 Amino acid determination

The amino acid profile was determined according to the method of Wang, L et al. (2019) [131]with some modifications. The minced chicken sausage (0.2 g) or Ab powder was hydrolyzed using 6 mol/L HCl (10mL) for 4h at 145±2 °C, filtered with a glass filter and transfer to a 50 mL volumetric flask and shaken well. The filtrate (10.0 μL) was concentrated using a rotary vacuum evaporator (Zhengzhou Kaixiang instrument equipment Co. Ltd., Henan, China) at 55 °C to remove the solvent. The liquid sample mixed with the derivatizing agent (20 μL) for 1 min, then mixed with another 20 μL of derivatizing reagent and sealed. The sample was transferred to an oven at 60 °C for 30 min. The amino acid content was analyzed using high-performance liquid chromatography (Huapu S6000, Huapu scientific instrument Technology Co., Ltd, Beijing, China). The measurement conditions were as follows: AccQ-Tag Ultra C18-250mm column, detection wavelength: 360 nm, column temperature: 25 °C, sample temperature: 20 °C, and flow rate: 1.2 mL/min.

2.2.21 Free fatty acids determination

The minced chicken sausage (0.1-10 g), which fat content was about 100-200mg, was mixed with pyrogallic acid (100mg), 95% ethanol (2ml), and water (4ml) in a 50ml centrifuge tube for the 5min. Next, 8.3 mol/L hydrochloric acid solution (10 mL) was added to a centrifuge tube, and the tube was shaken for 3 minutes. Then the centrifuge tube was put into a water bath at 70-80°C for hydrolysis for 40min, with shaking every 10min, taken out, and reduced temperature to room temperature after the hydrolysis was completed. The hydrolyzed sample was mixed with 95% ethanol (10 mL). Then the hydrolyzate was transferred to the fat extraction tube.

A 1:1 mixture of ether and petroleum ether (50ml) was added to the extraction tube, and the tube was shaken repeatedly 15 times to extract fat followed by a 15-minute break, and then the upper solution was poured into a 250ml conical flask. The above steps were repeated three times, and the upper extract was collected in a 250ml conical flask. The collected upper extract was concentrated using a rotary vacuum evaporator (Zhengzhou Kaixiang instrument equipment Co. Ltd., Henan, China) to remove the solvent. The residue was the fat extract. The fat extract was mixed with 8 mL of 2% sodium hydroxide methanol solution, the mixture was refluxed in a water bath at 80 °C for 30 min, and the sample was shaken every 5 min. Next, 15% methanol solution of boron trifluoride (7mL) was added in from the upper end of the condenser, and the temperature of the sample was rapidly lowered to room temperature after continuing to reflux for 2min.

10 ml n-heptane was accurately added followed by shaking for 2min, then a small

amount of saturated sodium chloride aqueous solution was added and stratified in a static position. The upper solution (5ml) was dehydrated with 3-5g anhydrous sodium sulfate and its free fatty acid was analyzed using a gas chromatograph (GC-Agilent7890B, Agilent Technologies, Santa Clara, CA, USA) according to the conditions reported by Barros et al.[135]. Three determinations were carried out for each treatment.

2.2.22 Thiobarbituric acid reactive substance (TBARS) assay

According to Xiong's method with some modification[136], 5 g of the ground sample and 20 ml of distilled water were homogenized with a homogenizer (IKA-T25, IKA Instruments Ltd, Staufen, Germany) for 3 minutes. 25 ml of trichloroacetic acid (TCA) aqueous solution was incorporated. The mixture stayed at room temperature for 1 h and then centrifuged at 2000 r/min for 10 min. The supernatant was diluted to 50 ml with distilled water. The diluent (5ml) was mixed with 0.02mol/l 2-thiobarbituric acid (TBA) aqueous solution (5ml). The mixture was incubated at 95 °C for 20 min before being cooled for 5 min at 0 °C in sequence. Then 10mL chloroform was added in. The mixture was centrifuged at 2000 r/min for 10 min. The absorbance of the supernatant was measured at 532 nm by the spectrophotometer against a distilled water blank. The TBARS value was expressed as mg/100 g sausage according to the following formula:

$$\text{TBARS value (mg/100 g)} = A532 \times 7.8 \quad (4)$$

2.2.23 Microbiological analyses

The microbiological analysis was performed on days 0, 5, 10, 15, 20, 25, 30, and 35 of storage at 4°C using standard procedures of China [137]. Total viable counts (TVC) were counted after incubation and results were expressed as CFU/g of sausage sample.

2.2.24 Statistical analysis

Data were analysed using analysis of variance (ANOVA) and expressed as mean ± standard deviation. The differences between means were considered significant at $P < 0.05$. Duncan's multiple range test was used to compare the means and identify significant differences between samples using SPSS Version 20.0 for Windows (IBM, Armonk, NY, USA)

2.2.25 Experimental design method

(1) The research of the chicken batters systems with Ab

① The effect of the Ab addition on the quality of chicken batters

The chicken batters were prepared according to the method in 2.2.3. The formulations of chicken batter were shown in Table 2.1. The samples without Ab powder were used as control. The chicken batters of 10 treatments were manufactured in each batch and 3 independent batches were prepared on the same day. The pH value, emulsion stability, color, and microstructure of chicken batters were determined, and the effects of Ab addition on the quality of chicken batters were investigated.

Table 2.1 – Chicken batters formulations with various fat and Ab levels (units: g/100 g)

Raw material/ Ingredients	Fat replacement level (%)									
	0%	2%	4%	6%	8%	10%	20%	30%	40%	50%
Meat batter										
Chicken meat	60	60	60	60	60	60	60	60	60	60
Back fat	20	19.6	19.2	18.8	18.4	18	16	14	12	10
Ab powder	0	0.4	0.8	1.2	1.6	2	4	6	8	10
Ice water	20	20	20	20	20	20	20	20	20	20
Total	100	100	100	100	100	100	100	100	100	100
Others (% of meat batter)										
Refined salt	1.4	1.4	1.4	1.4	1.4	1.4	1.4	1.4	1.4	1.4
Sodium tripolyphosphate	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Sugar	0.65	0.65	0.65	0.65	0.65	0.65	0.65	0.65	0.65	0.65
Ground white pepper	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15

② The effect of the addition method of Ab on the quality of chicken batters

Ab mushroom powder is mixed with soybean oil or water in 1:2 24 hours in advance, so that Ab powder can fully absorb water, oil and expand. The chicken batters were prepared according to the method in 2.2.3. The formula of chicken batter is shown in Table 2.2. One part of raw meat batters was stored in plastic bags at 4 °C for the analysis of rheological properties within 8 h. The other part was loaded in 50 ml polypropylene tubes and weighed. Next, the batters were heated to 80 °C for 30 min in a water bath, followed by cooling in ice water for 20 min to obtain chicken gel (cooked batters). The prepared chicken gels were stored at 4 °C until further analyses. The cooking yield (CY), water holding capacity (WHC) and color, TPA, dynamic rheological properties, LF-NMR spin-spin relaxation (T2), and microstructure of chicken batters were determined, the effects of Ab alone or in combination with soybean oil or water as fat substitutes on gel properties, rheology, water distribution, and microstructure of chicken batters were investigated.

Table 2.2 – The formulations of Chicken batters (units: g/100 g)

Raw material/ ingredients	CK	Ab	Ab+O	Ab+W
Meat batters (100g)				
Chicken meat	60	60	60	60
Pork Back fat	20	16	8	8
Ice water	20	20	20	20
Ab powder		4		
Ab powder +Soybean oil			4+8	
Ab powder+ Ice water				4+8
Total	100	100	100	100
Others (% of meat batters)				
Refined salt	1.4	1.4	1.4	1.4
Sugar	0.65	0.65	0.65	0.65
Sodium tripolyphosphate	0.3	0.3	0.3	0.3
Ground white pepper	0.15	0.15	0.15	0.15

③ Effect of the particle size of Ab on the quality of chicken batters

Three kinds of under sieves, D_1, D_2 and D_3, were obtained. The particle sizes of Ab powder was measured by a laser particle size analyzer (BT-9300H, Sichuan KeYicheng Technology Co., Ltd, Chengdu, China). The mean diameters of particles of D_1, D_2 and D_3 were 118μm, 100μm and 34 μm, respectively. Then, Ab mushrooms powder was stored in polyethylene bags in a refrigerator (4°C).

Ab powder with different particle size was mixed with soybean oil in 1:2 24 hours in advance for fully absorbing water, oil, and expanding. The formula of chicken batters were shown in Table 2.3. The production method of chicken batters as described in 2.2.3 and Figure 2.1. One part of raw meat batters was stored in plastic bags at 4 °C for the analysis of rheological properties within 8 h. The other part was loaded in 50 mL polypropylene tubes and weighed. Next, the batters were heated to 80 °C for 30 min in a water bath, followed by cooling in ice water for 20 min to obtain chicken gel (cooked batters). The prepared chicken gels were stored at 4 °C until further analyses. The CY, WHC, pH value, color, TPA, Dynamic rheological properties, LF-NMR spin–spin relaxation (T2), microstructure of chicken batters were determined. The effects of the particle size of Ab on the quality of chicken batters were investigated.

Table 2.3 – The formulations of chicken batters with different Ab particle sizes (units: g/100 g)

Raw material/ ingredients	CK	D_1	D_2	D_3
Meat batter (100g)				
Chicken meat	60	60	60	60
Back fat	20	8	8	8
Ab powder（118 μm）	-	4	-	-
Ab powder（100μm）	-	-	4	-
Ab powder（34 μm）	-	-	-	4
soybean oil	-	8	8	8
Ice water	20	20	20	20
Total	100	100	100	100
Others (% of meat batter)				
Refined salt	1.4	1.4	1.4	1.4
Sodium tripolyphosphate	0.3	0.3	0.3	0.3
Sugar	0.65	0.65	0.65	0.65
Ground white pepper	0.15	0.15	0.15	0.15

④ The effect of Ab on the gel properties of chicken myofibrillar protein

The MP was diluted to a final concentration of 70 mg/mL using a buffer containing 0.6 mol/L NaCl and 50 mmol/L K_2HPO_4/KH_2PO_4 (pH 6.5). The Ab powder was added to the MP solution at mass percentages of 0% (CK, control), 1% (T30), 2% (T2), 4% (T4), 6% (T6), respectively, and then the blended MP solution was homogenized at 15,000 rpm for 30 s in a homogenizer (IKA-T25, IKA Instruments Ltd, Staufen, Germany). A part of the Ab-MP mixed solution was used for measuring dynamic rheological properties. The rest of the samples were divided into 10 mL beakers, each beaker was filled with 10 g of the Ab-MP mixture and then sealed with plastic wrap. Next, the samples were subjected to a thermal gelation process (80 °C, 20 min) followed by overnight storage at 4 °C. Then, the water holding capacity, gel strength, texture, LF-NMR T_2, microstructure, and secondary structure of the thermal gel were measured.

(2) The research of the recipe composition and quality properties of the sausages

① Optimization of potato starch addition in sausage

The raw materials for making sausages were first prepared, including 240g of chicken breast, 32g of pork back fat, 16g of Ab powder, 32g of soybean oil, 80mL of ice water, 1.2g of edible sodium polyphosphate, 0.6g of white pepper powder, 2.6g of white sugar, 5.6g of salt, 0.80g of chicken essence. The addition amount of tomato starch was 8g, 12g, 16g, 20g, 24g respectively (ie 2%, 3%, 4%, 5%, 6% of chicken batters). Five sets were prepared according to the method of 2.2.5 and Figure 2-2. The color, flavor, taste, texture and general acceptance of the sausages were graded respectively for screening out the best addition amount of potato starch in sausage.

② Optimization of chicken essence addition in sausage

The raw materials for making sausages were first prepared, including 240g of chicken breast, 32g of pork back fat, 16g of Ab powder, 32g of soybean oil, 80mL of ice water, 1.2g of edible sodium polyphosphate, 0.6g of white pepper powder, 2.6g of white sugar, 5.6g of salt. The addition amount of tomato starch was the optimal amount obtained in (1). The addition amounts of chicken essence were 0g, 0.4g, 0.8g, 1.2g, 1.6g respectively (ie 0%, 0.1%, 0.2%, 0.3%, and 0.4% of chicken batters). Five treatments of sausages with different recipes were prepared according to the method of 2.2.5 and Figure 2.2. The color, flavor, taste, texture and general acceptance of the sausage were graded respectively for screening out the best addition amount of chicken essence in sausage.

③ Optimization of refined salt addition in sausage

The raw materials for making sausages were first prepared, including 240g of chicken breast, 32g of pork back fat, 16g of Ab powder, 32g of soybean oil, 80mL of ice water, 1.2g of edible sodium polyphosphate, 0.6g of white pepper powder, 2.6g of white sugar. The addition amount of tomato starch and chicken essence was the optimal amount obtained in (1) and (2). The addition amounts of refined salt were 4g, 4.8g, 5.6g, 6.4g, 7.2g respectively (ie 1%, 1.2%, 1.4%, 1.6%, and 1.8% of chicken batters). Five treatments of sausages with different recipes were prepared according to the method of 2.2.5 and Figure 2.2. The color, flavor, taste, texture and overall acceptance of the sausage were graded respectively for screening out the best addition amount of refined salt in sausage.

④ Optimization of the premix of Ab and soybean oil addition in sausage

Ab powder was mixed with soybean oil in 1:2 24 hours in advance for absorbing oil and expanding. The raw materials for making sausages were first prepared, including 240g of chicken breast, 80mL of ice water, 1.2g of edible sodium polyphosphate, 0.6g of white pepper powder, 2.6g of white sugar. The addition amount of tomato starch, chicken essence, and refined salt was the optimal amount obtained in (1), (2), and (3). To replace

0%, 30%, 60%, and 90% of pork back fat, respectively, the addition amounts of the premix of Ab and soybean oil were 0g（CK）, 24g（T30）, 48g（T60）, 72g（T90）, respectively (ie 0%, 6%, 12%, and 18% of chicken batters), the corresponding additions of pork back fat were 80g, 56g, 32g and 8g, respectively. Four treatments of sausages with different recipes were prepared according to the method of 2.2.5 and Figure 2.2. The color, flavor, taste, texture and overall acceptance of the sausage were graded respectively for screening out the best addition amount of the premix of Ab and soybean oil in sausage.

⑤ Evaluation of the quality characteristics of the sausage

The formula of chicken sausages as shown in Table 2.4. Four formulations were designed to reduce fat (0%, 30%, 60%, and 90% reductions) by the addition of Ab and soybean oil premix. The preparation method of chicken sausages as described in 2.2.5 and Figure 2.2. Next, the sausages were stored at 4℃ until the laboratory analysis during the 35-days storage period. Chemical composition, amino acid, and fatty acid profile were evaluated only on day 0. The sausages were placed at room temperature for 30 min in advance for the microbial analysis. Analyses of other indicators for each formulation were conducted at 0, 5, 10, 15, 20, 25, 30, and 35 days of storage in triplicate.

Table 2.4 – Formulations of chicken sausages with Ab powder

Raw material/ ingredients	Formulations(%)			
	CK	T30	T60	T90
Meat batter				
Chicken meat	60	60	60	60
Back fat	20	14	8	2
Ab powder	0	2	4	6
Soybean oil	0	4	8	12
Ice water	20	20	20	20
Total	100	100	100	100
Others (% of meat batter)				
Refined salt	1.4	1.4	1.4	1.4
Sodium tripolyphosphate	0.3	0.3	0.3	0.3
Chicken essence	0.1	0.1	0.1	0.1
Sugar	0.65	0.65	0.65	0.65
Ground white pepper	0.15	0.15	0.15	0.15
Potato starch	5	5	5	5

2.3 Technical route figure

A detailed research plan was developed in order to define the scope, objectives and content of the research and to plan the entire research process specifically so that the study could be implemented as planned and thus successfully complete the research task (Fig. 2.2). To develop a new type of low-fat chicken sausages using Ab, it is necessary to determine the optimal amount and method of addition of Ab, the particle size of Ab, the formulation of low-fat chicken sausages, and evaluate the nutritional and storage properties of the sausages, the economic benefits and the effects of application in enterprises. The research results will provide a basis for the development of a new technology for producing low-fat chicken sausage using Ab. The study is divided into five stages.

The first stage is the theoretical analysis. The main areas of focus are the following. Analysis of the impact and consumption trends of meat and meat products, analysis of the characteristics of low temperature meat products and progress in fat reduction technology, analysis of the current status of cultivated mushroom production and its application in food processing, and analysis of the health properties and application prospects of Ab in low fat meat products.

The second stage is to define the main objectives and tasks of this study based on the theoretical analysis.

For the purpose of the study, the third phase defines the content of the experiment. The experiment included two main parts. One part investigated the effect of Ab on the quality of the minced chicken system. The research was carried out in four aspects： selecting the optimal amount of fat replacement in chicken batters by Ab, selecting the optimal way to replace fat in chicken batters by Ab, selecting the optimal particle size of Ab and exploring the mechanism of Ab to improve the gelation properties and water retention capacity of low-fat chicken batters. Through these four aspects of research, a method for obtaining high quality low-fat chicken batters containing Ab is established and the feasibility of applying Ab to low-fat chicken batters is theoretically determined. The other part was the production of low-fat chicken sausages using low-fat chicken batters. Three aspects of the study were conducted: determining the optimum formulation composition for the production of low-fat chicken sausages containing Ab, determining the technological solutions for the production of low-fat chicken sausages containing Ab, and investigating the nutritional and storage characteristics of low-fat chicken sausages

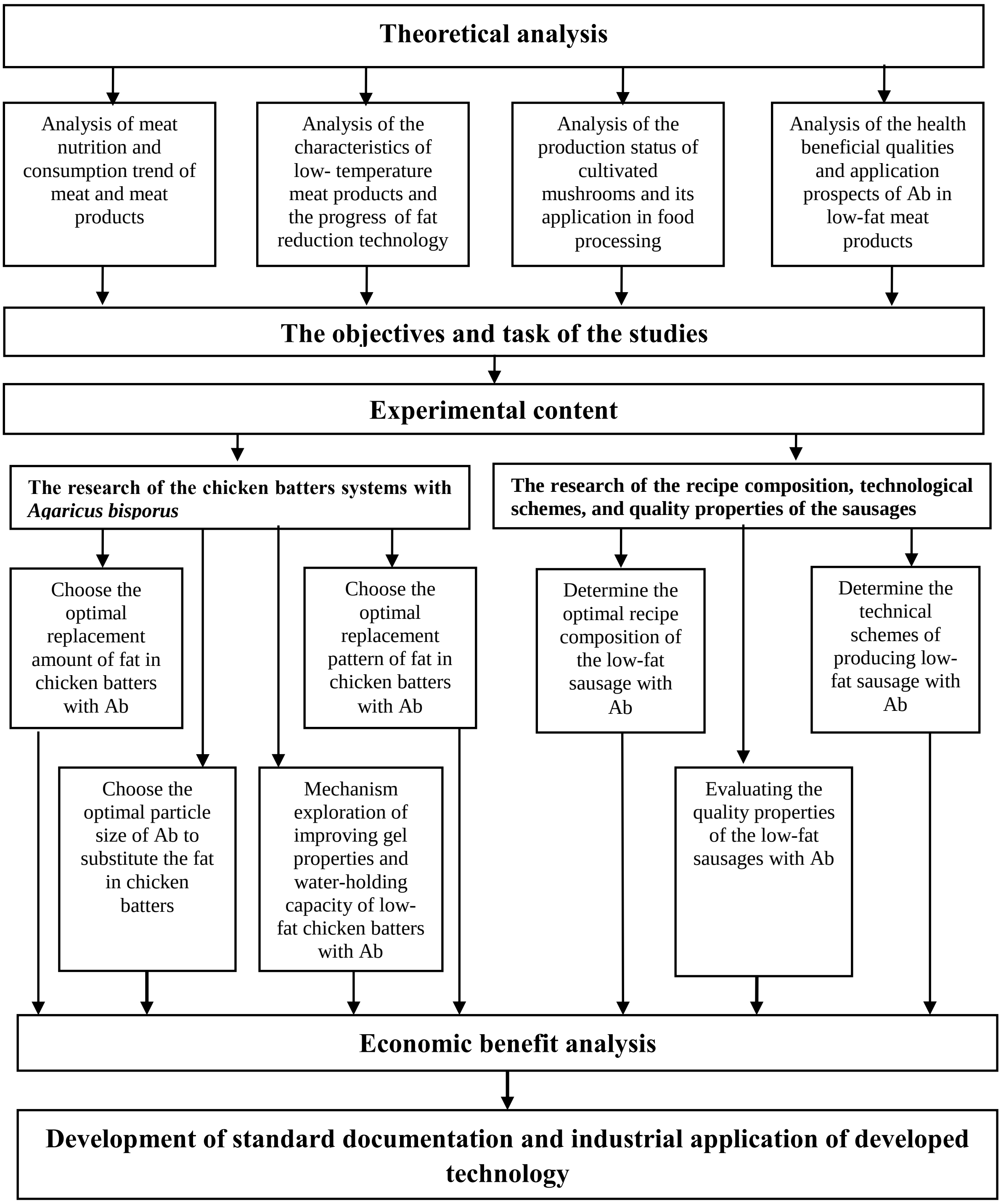

Figure 2.2 – Technical route

containing Ab. Through these three studies, the composition of the low-fat chicken sausage recipe, the production technology, the quality characteristics and the shelf life will be determined.

The fourth stage is the economic benefit analysis. The economic benefits of producing low-fat chicken sausages containing Ab in food processing enterprises were analysed to assess the expected economic benefits of implementing the results of this study in enterprises and to determine the feasibility and prospects of the practical application of the research results.

The fifth stage is the implementation of the research results in practice. A standard document on the production processing and operational key points of low-fat chicken sausages containing Ab was developed and the research results were approved by the industry under factory production conditions at Henan Jiaduoduo Foods Co Ltd (China) and Henan Fengxiangfengwei Food Co Ltd (China).

The experimental part of the thesis was conducted in the laboratories of Food Technologies Faculty of Sumy National Agrarian University (Sumy, Ukraine); “National Pork Research and Development Technology Center” of Henan University of Science and Technology (Xinxiang, China); “Intensive Processing and Quality Control of Agricultural Products” Key Laboratory of Henan Province's universities (Xinxiang, China), and “Functional Food and Ingredients” Key Laboratory of Xinxiang City (Xinxiang, China); Institute of Structural Biology, Nanyang Technological University (Singapore).

CONCLUSION OF SECTION 2

1. The methods used in the thesis work are identified, including a variety of methods such as theoretical studies, raw materials and ingredients, experimental methods, practical testing and technology dissemination.
2. Established a Technical route for developing low-fat chicken sausage with Ab mushroom.

SECTION 3 THE RESEARCH OF THE CHICKEN MEAT BATTERS SYSTEMS WITH AB

3.1 The effect of the Ab addition on the quality of chicken batters

Fat has an important influence on the texture, water retention and flavor of emulsified meat products. When the fat content is lowed, the quality of meat products will be severely degraded, so it is difficult to develop low-fat meat products. When replacing fat with Ab, the quality of meat products will be affected by the amount, adding method, and particle size of Ab. However, at present, there is no relevant research report on the application of Ab to low-fat chicken products. Therefore, it is necessary to study these aspects.

The gelation of protein is an important part of the functional properties of meat products, and is mainly responsible for the functional properties of processed meat products, including firmness, elasticity, chewiness, and cohesiveness. Muscle proteins are composed of approximately 60% salt-soluble myofibrillar proteins, which can form three-dimensional gels upon heating, and the effects of these proteins on gel formation, water retention, structure, and rheological properties have been widely reported. It is crucial to study the properties of mixed gel of myofibrillar protein and fat substitute to judge the quality of meat products, which can provide theoretical guidance for the development of low fat meat products. Therefore, it is necessary to study the properties of the composite gel of Ab and chicken myofibrillar protein, so as to provide guidance for the application of Ab chicken emulsified products theoretically.

3.1.1 pH

As can be seen in Figure 3.1, the pH value of chicken ground meat significantly increased with the increase in the amount of fat replacement. This result was consistent with previous studies. Jeehwan Choe and Juri Lee(2018) observed an increase in the pH of pork batter with the addition of winter mushroom powder[129]. This might be due to the buffering effect of proteins in Ab powder.

Ab contains high levels of basic amino acids such as arginine and histidine at a level of 99.8μmoL and 133.7 μmoL per 100 g sample on a wet weight basis[138], the pH value of Ab was 6.50[139],which was much higher than that of the control(5.99). Higher pH value was more conducive to the formation of meat gels, resulting in a finer gel structure and greater gel strength[140]. This result was inconsistent with the report of Wang, Cheng Li et al.(2019), who reported that the replacement of pork back fat with fried *Pleaurotus eryngii* resulted in a reduction in the pH of the sausage, mainly because the pH of fried *P. eryngii* was lower than that of pork back fat [131].

Based on the above analysis, adding Ab powder increased the pH value of chicken batters, which would be conducive to improve the gel structure of meat paste.

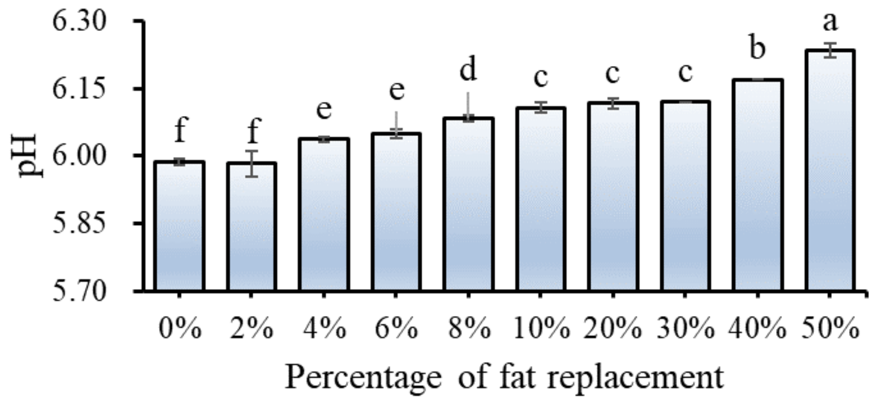

Figure 3.1 – pH value of chicken batters with Ab powder as fat substitution

Note: a–f Means with different letters are significantly different (p<0.05).

3.1.2 Emulsion stability

The change trend of cooking loss, water loss and fat loss of samples with different fat replacement percentage were different (Table 3.1).The amount of cooking loss and water loss decreased with the increase of fat replacement amount, fat loss fluctuated with the increase of fat replacement amount and reached the maximum at 10% replacement (Table 3.1). The cooking loss and water loss were significantly ($P < 0.05$) different from those in the control group when the fat replacement amounts were more than 2% and 8% respectively, which was closely related to the increase in pH of the chicken batters with different fat replacement amounts. Because the high pH value of chicken batters would have increase the net negative charge of chicken protein, which could have increased its water holding capacity and, subsequently, reduced cooking loss and water loss[141]. In addition, Ab contain hemicellulose, cellulose and lignin at a level of 10.10% ,11.39%, and 14.68% respectively[139], which have oil absorption and hydration (water uptake, retention, and swelling) properties[142, 143]. This result was consistent with the report of Minyi Han, et al. (2017) who found that adding dietary fiber significantly reduced cooking loss of meat products[142]. However, the results of fat loss in this study were different. One possible reason for this was that the water holding capacity of Ab was stronger than that of oil holding capacity, and a small amount of Ab preferentially adsorbsed water. With the increase of the amount of Ab powder, the content of free water in the system decreased, subsequently, Ab started to absorb a lot of oil, as a result, the loss of fat reduced.

Table 3.1 – Emulsion stability of chicken batters with different amounts of fat replacement

Percentage of fat replacement	Emulsion stability		
	Cooking loss (%)	Water loss (%)	Fat loss (%)
0	13.47±0.37a	12.32±0.15a	1.14±0.03d
2%	12.83±0.25a	11.77±0.08a	1.06±0.01de
4%	12.03±0.75b	10.89±0.03a	1.15±0.01d
6%	11.33±0.15b	10.12±0.05a	1.21±0.02c
8%	9.87±0.13c	8.33±0.18ab	1.54±0.02bc
10%	8.66±0.45d	6.79±0.07bc	1.87±0.01a
20%	7.52±0.36e	5.74±0.05cd	1.78±0.02bc
30%	5.60±0.18f	4.22±0.02cd	1.38±0.00bc
40%	3.47±0.31g	2.38±0.01d	1.08±0.01de
50%	1.7±0.30h	0.97±0.01e	0.73±0.01e

Note: All values are mean ± SD. a–g Means within a column with different letters are significantly different ($p<0.05$).

3.1.3 TPA

According to Table 3.2, the texture parameters of chicken batters increased with the increase of the amount of fat replaced by Ab, but the springiness, cohesiveness and chewiness decreased when the added amount was high. When the fat replacement amount was more than 10%, the hardness began to increase significantly. When the amount was 50%, the hardness was the highest, which was 1.37 times of the control. The springiness of the samples with 8% to 20% fat replacement was significantly higher than that of the control group and other experimental groups. When the fat replacement was 10%, the springiness reached the maximum value of 0.916, but there was no significant difference among the three sample groups. The samples of 40% and 50% group deteriorated in springiness, which was consistent with the research results of Jung Ha et al.(2001) that adding Ab powder to surimi increased the hardness and springiness of surimi [144]. However, Wong km et al. （2017） reported that adding wet Ab to ground beef (80 / 20 fat blend) reduced the hardness of beef [145]. Chung SI et al.(2010) found that adding King Oyster mushroom to cutlefish meat paste reduced the hardness of minced meat [116], which were inconsistent with the present results. The possible reason was that King Oyster Mushroom with 87.1% moisture content, leading to a reduction in the hardness of surimi.

When the replacement amount was more than 10%, the chewiness of the sample group was significantly higher than that of the control group. When it was 30%, the chewiness was the best. Nevertheless, there were not the significant difference between 20%, 40% and 50% sample groups. Ab powder did not have a strong effect on improving the cohesiveness of chicken batters. The cohesiveness of the samples with 2% to 8% of fat replacement increased slightly, while only the samples with 6% and 8% of fat replacement were significantly higher than the control. When the amount of fat replacement was greater than 8%, the cohesiveness was significantly worse than that of the control. This result was consistent with that reported by Chung Si et al. (2010)[116].

The results of this experiment were contrary to the results of refined dietary fiber from ascidian on the texture of surimi. Hong-Sun Yook et al. (2000) reported that fish paste prepared with refined dietary fiber from ascidian, with the increase of fiber content, the springiness decreased and the viscosity increased [146]， which might be due to the different quality of mushrooms, raw materials and meat.

In the meat processing, high ionic strength was more conducive to the extraction of myofibrillar protein in meat [147], the structural characteristics of cooked meat products were closely related to the gelation of myofibrillar proteins [127, 148]. Ab contain higher Ca, K, Mg, Na and other elements, the contents were 860, 38105, 11099, 234 mg kg^{-1} dm, respectively [149]， and high metal elements increased the ionic strength of chicken meat, which was conducive to the extraction of soluble protein from chicken meat, thus form meat gel structure. Another reason for the gelation of meat emulsion structure might be related to the presence of dietary fiber and its water-holding behavior [150]. The rich dietary fiber of Ab was beneficial to the formation of gel structure [139].

High addition of Ab was detrimental to springiness, cohesiveness and chewiness, and this inhibitory effect could be attributed to the presence of other components in the mushroom. Such as ash, which will adversely affect the interaction between the hydrophilic groups of the protein and water molecules or the protein-protein interaction [151].

Based on the above analysis, the hardness, springiness and chewiness of chicken batters with 20% fat replacement improved significantly, although the cohesiveness decreased. Therefore, 20% replacement was considered the best level.

Table 3.2 – The textural profile of cooking chicken batters with different amounts of fat replacement

Percentage of fat replacement	Firmness(g)	Springiness	Cohesiveness	Chewiness(g)
0%	7392.02±462.48d	0.889±0.006bcd	0.71±0.02c	4748.94±310.54c
2%	7481.99±293.28d	0.901±0.009abc	0.72±0.01bc	4862.28±637.47c
4%	7541.56±259.89d	0.903±0.018abc	0.72±0.01bc	4913.39±107.87c
6%	7565.70±650.23d	0.908±0.028ab	0.73±0.01ab	4993.27±372.69c
8%	7617.12±368.64d	0.910±0.018a	0.73±0.01a	4929.70±414.02c
10%	7730.49±158.19d	0.916±0.019a	0.70±0.00d	4976.29196.75c
20%	8380.79±350.12c	0.911±0.003a	0.69±0.01e	5569.27±302.43b
30%	8610.34±287.88bc	0.908±0.002ab	0.67±0.01f	6256.09±209.74a
40%	9042.53±312.73b	0.883±0.010cd	0.66±0.01g	5712.12±260.60b
50%	10090.66±473.84a	0.874±0.006cd	0.65±0.00g	5725.51±298.78b

Note: All values are mean ± SD. a–g Means within a column with different letters are significantly different ($p<0.05$).

3.1.4 Color

The addition of Ab powder significantly ($P < 0.05$) changed the L*, a*, b* values and whiteness of chicken batters (Table 3.3). The L* value and whiteness decreased significantly ($P < 0.05$) and lower than control group with the increase of Ab powder content. Meanwhile, the value of a* and b* increased significantly ($P < 0.05$) and higher than control group. The possible reason was that L* value and whiteness of Ab powder were low. Nevertheless, a* and b* value of chicken batters were increased, which may be due to the yellowish pigmentation in Ab powder. This result was consistent with the report of Jung UK ha et al. (2001) who found that adding Ab powder reduced the L* value and increased the a* value of surimi [144]. Arun K. et al. (2010) reported that the color index of low-fat chicken nuggets increased with the incorporation of apple pulp level. The high redness and yellowness of apple pulp could be the reason for the increased chroma index of the product [152].

Nevertheless, the results of this study were not completely consistent with the results of Ceron-Guevara et al. (2019) [153] who reported that the addition of Ab powder had a significant (*$P < 0.05$*) effect on the L* value, but not on the a* and b* values of beef patties. The possible reason was that the beef had higher a* and b* values than chicken.

The addition of Ab powder significantly reduced the whiteness of chicken batters. When the amount of mushroom replacing fat was 50%, the whiteness value was close to that of pork sausage with monascus color [131], but no pigment was added in this experiment. Therefore, Ab powder could also be used as a natural food pigment, reducing the amount of other pigments.

Table 3.3 – The color of cooking chicken batters with different amounts of fat replacement

Percentage of fat replacement	L*	a*	b*	Whiteness
0%	85.16 ± 0.21a	0.85 ± 0.01h	8.81 ± 0.08h	82.72± 0.22a
2%	81.36 ± 0.28b	1.06 ± 0.05f	10.58 ± 0.06g	78.54± 0.22b
4%	79.80 ± 0.25c	0.74 ± 0.01i	12.12 ± 0.06f	76.43± 0.19c
6%	77.79 ± 0.05d	0.98 ± 0.02g	12.17 ± 0.10ef	74.66± 0.07d
8%	77.69 ± 0.46d	1.18 ± 0.03e	12.33 ± 0.08e	74.49± 0.38d
10%	74.48 ± 0.47e	1.20 ± 0.04e	13.11 ± 0.19d	71.28± 0.34e
20%	68.82 ± 0.11f	1.78 ± 0.08d	17.48 ± 0.17c	64.21± 0.17f
30%	64.90 ± 0.33g	2.36 ± 0.05c	18.17 ± 0.11b	60.41± 0.25g
40%	61.15 ± 0.16h	3.0 ± 0.02b	18.19 ± 0.00b	57.00± 0.15h
50%	60.17 ± 0.32i	3.28 ± 0.03a	18.50 ± 0.09a	55.96± 0.30i

Note: All values are mean ± SD. a–i Means within a column with different letters are significantly different ($P < 0.05$).

3.1.5 Scanning electron microscopy

The control group and the sample with 20% fat replacement, which was considered to have the best qualitative improvement, were selected for SEM observation, as shown in Figure 3.2. Observation of these microstructure changes contributed to explain water-binding abilities and the textural properties of the cooked chicken batters.

The scanning electron microscope image reflected the microstructure of the meat emulsion gel, and could be used to explain the water holding capacity of the gel. Smooth gels could effectively bind water, while coarse gels were fragile and had poor water holding capacity [32]. The control group (as shown in Figure 3.2, A, C) without adding Ab powder had rough surface and large cavity structure. The pore in the microstructure of gel was water channe [154]，and large macropores made it easy for water to escape from the protein network, resulting in increased water loss.

The addition of Ab powder increased the gel network density of chicken batters, and the gel showed a compact, uniform and continuous appearance (as shown in Figure 3.2, B, D), the pores in the microstructure decreased as well as. The observation of microstructural changes showed that Ab powder as a fat substitute could improve the water retention of chicken batters, fill the protein matrix and improve the gel strength of chicken batters, which were consistent with the results of water loss and tissue profile in present study. It is reported that the addition of regenerated cellulose could effectively reduce the pores of water channels and fill the protein matrix, which was easy to intercept the water [155]. It was reported that the cellulose content of Ab powder was as high as 21.49%[139], so it could be inferred that Ab powder prevented water from seeping out through the capture and retention of moisture in the muscle protein matrix and reduced the water channel through the filling action, thereby densified the gel network. Therefore, adding Ab could improve the water holding capacity and microstructure of chicken batters.

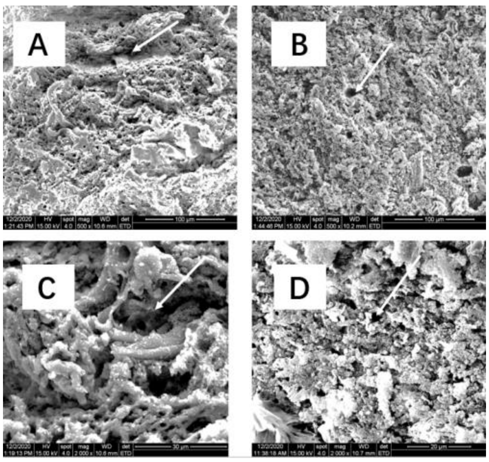

Figure 3.2 – The microstructures of cooked chicken batters observed by scanning electron microscopy.

Note: (A) Control without Ab, 500 x visual fields; (B) Replace 20% of the fat with Ab, 500 x visual fields ; (C) Control without Ab, 2000 x visual fields; (D) Replace 20% of the fat with Ab.

3.2 The effect of the addition method of Ab on the quality of chicken batters

3.2.1 Cooking yield (CY), water holding capacity (WHC) and color

Generally, both CY and WHC can indicate the quality and moisture-binding capacity of meat batter. Table 3-4 shows the CY, WHC, and color of the meat batters with or without fat substitutes. The CY and WHC of the samples containing fat substitutes were significantly increased ($P < 0.05$) compared to CK, indicating that the three fat substitutes significantly improved the moisture-binding capacity of meat batter. The increase in CY and WHC might be due to several components such as cellulose, hemicellulose, and lignin present in Ab [139], which show good oil-absorbing and water-binding properties [142, 143]. However, there were no significant ($P < 0.05$) differences between the samples of Ab and Ab+O in CY and WHC in the present study. A possible explanation for this result in our study could be due to the presence of less pork back fat in the sample of Ab+O than Ab.

The color characteristics of meat products are essential for the consumers' acceptance of the product [156]. Compared to CK, the values of L* and whiteness of the samples containing fat substitutes decreased significantly ($P < 0.05$), while a* and b* value were increased significantly ($P < 0.05$). This outcomes may be related to the color of Ab powder, which is low in L* and whiteness values, and high in a* and b* values, due to its characteristic yellowish pigmentation[153]. These results of L* and b* values were consistent with the report that the addition of Ab powder reduced the L* value and increased the b* value of beef paste [157]. Furthermore, it has reported that the color index of low-fat chicken nuggets containing apple pulp increased with the level of apple pulp, which exhibited high redness and yellowness [152]. In addition, the whiteness, L*, a*, and b* values of cooked batters treated with Ab+O was significantly ($P < 0.05$) different from the treatment of Ab and Ab+W, which might be attributed to the color of soybean oil and the difference in solubility of the mushroom pigments in water and oil [158, 159].

Table 3.4 – CY, WHC and color of chicken batters without or with fat substitutes

Sample	CY	WHC	Whiteness	*L**-value	*a**-value	*b**-value
CK	85.01±1.19^{c}	85.12±0.82^{c}	78.70±0.17^{a}	83.35±0.12^{a}	1.50±0.06^{c}	13.21±0.07^{c}
Ab	90.60±0.94^{a}	88.07±1.31^{a}	59.62±0.09^{c}	63.34±0.09^{c}	1.97±0.11^{a}	16.54±0.08^{a}
Ab+O	89.62±0.47^{a}	87.79±1.13^{a}	63.87±0.12^{b}	67.57±0.15^{b}	1.78±0.08^{b}	15.87±0.10^{b}
Ab+W	87.55±0.81^{b}	86.88±0.30^{b}	60.07±0.29^{c}	63.71±0.21^{c}	1.97±0.14^{a}	16.56±0.12^{a}

Note: CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% Ab powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% Ab powder + 40% water; CY: cooking yeild; WHC: water holding capacity. Different letters (a–c) in the same column indicate significant differences ($P < 0.05$) between samples.

3.2.2 TPA

Figure 3.3 shows the texture profile of chicken gel with or without Ab powder. Compared with the CK, the addition of Ab powder significantly ($P < 0.05$) improved the hardness, springiness, and chewiness, and reduced the cohesiveness of the batters. The addition of Ab powder and soybean oil significantly ($P < 0.05$) enhanced the hardness, springiness, and cohesiveness, and exhibited no significant effect on the chewiness of the chicken gel. The addition of Ab powder and water significantly ($P < 0.05$) increased the hardness, springiness, and chewiness, and exhibited no significant effect on the cohesiveness of the batters. Thus, it could be concluded that the combination of Ab powder and soybean oil or water significantly ($P < 0.05$) improved the texture characteristics of the batters. These results are supported by a previous study, wherein it was observed that the rich dietary fibre from Ab was beneficial for the textural properties of cooked beef emulsion [139]. The structural characteristics of cooked meat products were closely related to the gelation of myofibrillar proteins [127, 148]. During the preparation of meat batters, high ionic strength is helpful for the extraction of myofibrillar protein from meat [147]. Ab powder is rich in Ca, K, Mg, Na, and other elements, which increases the ionic strength of chicken batters and makes it conducive to extract salt-soluble proteins from chicken [149] , proving it beneficial for the formation of the three-dimensional structure of heat-induced gel [160]. The Ab+O sample had the highest springiness and cohesiveness, suitable hardness, and similar chewiness to the control group, thus showing the best texture attributes among all the samples. This could be due to the fact that the premixing of Ab powder and oil disperses oil droplets in Ab particles, forming smaller oil droplets than fat globules, reducing the accumulation of oil droplets and the interfacial tension between water and oil phases, and accelerating the emulsification process of meat. This result agrees with the results of Choi et al. (2009) who reported the effects of vegetable oils and rice bran fiber on the textural properties of low-fat meat emulsion systems[161].

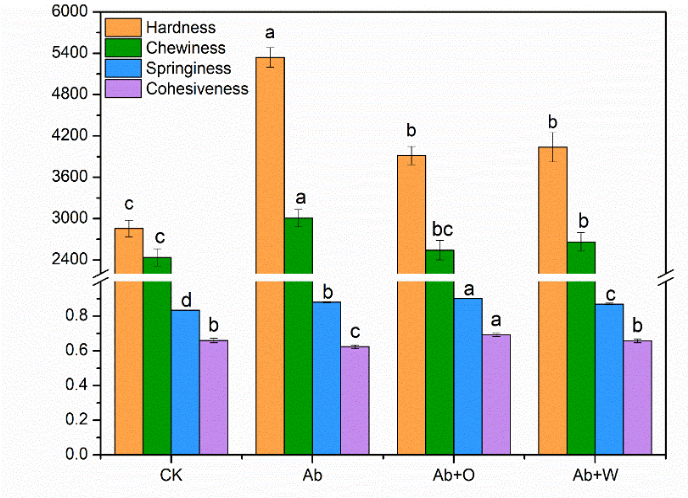

Figure 3.3 – The textural attributes of cooked chicken batters without or with fat substitutes.

Note: CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% Ab powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% Ab powder + 40% water. Values bearing different superscripts above the bar with the same color indicate significant differences ($P < 0.05$) between samples.

3.2.3 Dynamic rheological properties

Dynamic rheological properties reflect the continuous changes in myofibrillar protein and protein-protein interactions (especially the myosin protein), which are closely related to the intramolecular and intermolecular binding in protein molecules [162]. The storage modulus (G') indicates the changes in the elastic properties of the gel network structure and matrix strength [163]. Figure 3.4 shows the elasticity of chicken batter during heating by measuring the storage modulus (G').

The curve of G' of all samples showed similar characteristics with increasing temperature (Figure 3.4). There was a typical initial increase until the peak was reached at about 54.52 °C, which suggested that the myosin head had started aggregating and initiated the protein denaturation, resulting in the formation of weak gel network structures [154]. Subsequently, G′ markedly decreased and reached its peak value at

62.5 °C, which might be related to the uncoiling of the myosin tail leading to an increase in protein mobility and destruction of the protein network that had previously formed at a lower temperature [164]. The G' value then sharply increased because of the formation of new chemical bonds and increased cross-linking between the proteins, causing the viscous sol to simultaneously transform into an elastic gel network. The rheological results of our study were in accordance with the typical characteristics of meat emulsion [162].

The final G' values of the three samples with fat substitutes were higher than that of the control samples, indicating that these substitutes could improve the elastic properties of the gel network structure and matrix strength of the chicken gelling system. This could be attributed to the rich dietary fibre from Ab powder, which might act as a filler and dehydrator, or change the protein structure, thereby enhancing the gel structure. It has reported that dietary wheat could function as a dehydrating agent in surimi and change the tertiary structure of the protein in the sol phase, as reflected in the alterations of the hydrophobic side chain environment, making them more solvent-exposed. After heating, it resulted in hydrophobic contact between protein and fibre and an effortless nonspecific solidification, thereby forming a continuous, compact, and dense uniform structure in batter [165].

Among all the treatments, the sample of Ab+O showed the highest G' value. In contrast, the sample of Ab+W displayed the lowest G' value, indicating that Ab+O showed the best ability in improving the meat batter elasticity than Ab and Ab+W. This was in agreement with the findings of the texture profile in this study. One possible explanation for the improved elasticity of chicken batter is that the premixing of Ab powder with soybean oil is similar to oil drop pre-emulsification. Many studies have confirmed that the addition of pre-emulsified vegetable oil could improve the G' level of meat batters [33, 161].

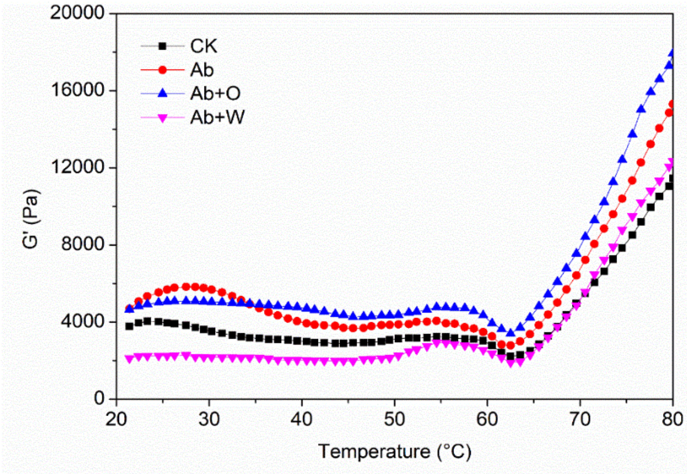

Figure 3.4 – The storage modulus (G') of chicken batters without or with fat substitutes.

Note: CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% Ab powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% Ab powder + 40% water. Values bearing different superscripts above the bar with the same color indicate significant differences ($P < 0.05$) between samples.

3.2.4 LF-NMR spin-spin relaxation (T2)

The low-field pulse NMR technique measures the T2 relaxation time and estimates the fluidity and structural properties of the components of the water molecules partially fixed by the proteins in the gel system.

Figure 3.5 and Table 3.5 show the continuous distribution of NMR T2 relaxation time and three T2 relaxation times of water protons and corresponding peak area fractions of water in cooked chicken batters. Three separate peaks were identified as T2b, a fast minor component (0–3% of total water), T21, a major component (91–98% of total water), and T22, a slow component (1–6% of total water) with the relaxation time of 0–1 ms, 40–60 ms, and 300–900 ms, respectively. In general, T2b reflects water molecules tightly bound or closely connected to myofibrillar proteins through strong hydrogen bonds [166]. T21 corresponds to immobilized water trapped within the myofibrillar protein matrix and tightly bound to monolayer water molecules [167]. T22 is related to free water that is weakly bound to the gel system and is the most fluid water component in the gel[168]. There were no significant ($P < 0.05$) differences in T2b of all samples, while the PT2b of Ab+O was significantly ($P < 0.05$) smaller than that of the other three samples, suggesting that the addition of Ab powder and soybean oil changed the bonding state of water and protein in chicken gel. It has reported that bamboo shoot dietary fibre did not influence the proportion of bound water in pork meat batter, which was consistent with the results of the samples of Ab and Ab+W in present experiment [169]. Compared to CK, the T21 values significantly decreased in the samples of Ab and Ab+O ($P < 0.05$), indicating that Ab powder or the mixture of Ab powder and oil could significantly ($P < 0.05$) enhance the gel's ability to bind to water and weaken the freedom degree of moisture. A significant ($P < 0.05$) decrease in T22 was observed in the gel system of chicken batter of Ab and Ab+O, suggesting that the addition of Ab powder or mixture of Ab powder and soybean oil decreased the water mobility of chicken meat gel and increased CY and WHC of chicken batters.

The amount of immobilized water in the Ab and Ab+O samples was significantly ($P < 0.05$) higher, while the amount of free water was significantly lower than those of the other two groups. The variation trend of T21 and PT21 between Ab and Ab+O was inconsistent. However, it has been reported that specific changes or trends between T21 and PT21 might be incompatible [154, 170]. The incompatibility between T21 and PT21 might be ascribed to the fact that the chicken batters with Ab powder and soybean oil formed a denser gel structure. The smaller the gel pore, the smaller the degree of freedom of the water wrapped by the myofibrillar protein matrix. A part of the free water in cooked chicken batters could have transformed to immobilized water, which may be related to the gel network structure.

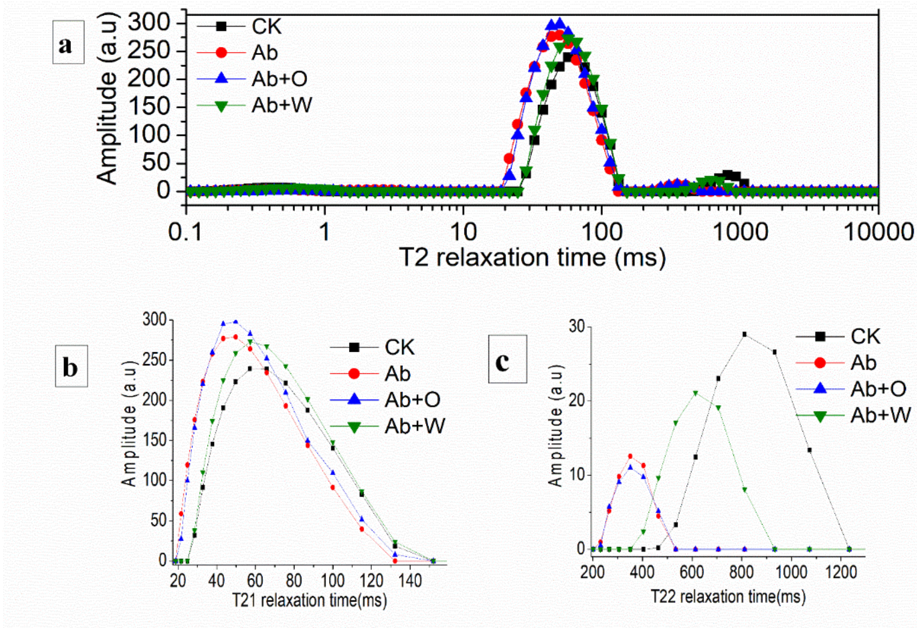

Figure 3.5 – The continuous distribution of NMR T2 relaxation time of chicken gel without or with fat substitutes.

Note: T2 relaxation time of chicken gel (a). T21 relaxation time of chicken gel (b). T22 relaxation time of chicken gel (c). CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% Ab powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% Ab powder + 40% water. Values bearing different superscripts above the bar with the same color indicate significant differences (*P* < 0.05) between samples.

Table 3.5 – T2 relaxation times and corresponding peak areas percentages of water in cooked chicken batters

Samples	T_{2b} (ms)	T_{21} (ms)	T_{22} (ms)	PT_{2b} (%)	PT_{21} (%)	PT_{22} (%)
CK	0.43 ± 0.18^a	57.22 ± 3.04^a	811.13 ± 25.71^a	2.94 ± 0.56^a	91.61 ± 1.41^d	5.46 ± 0.17^a
Ab	0.43 ± 0.12^a	49.77 ± 2.15^b	351.12 ± 23.41^c	2.38 ± 0.45^a	95.82 ± 1.24^b	1.80 ± 0.43^c
Ab+O	0.57 ± 0.10^a	49.77 ± 3.23^b	351.12 ± 20.62^c	0.42 ± 0.12^b	97.92 ± 1.03^a	1.66 ± 0.25^c
Ab+W	0.65 ± 0.08^a	57.22 ± 2.01^a	513.59 ± 20.48^b	2.66 ± 0.36^a	93.24 ± 1.36^c	3.52 ± 0.36^b

Note: CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% A. bisporus powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% Ab powder + 40% water. Different letters (a–d) in the same column indicate significant differences (*P* < 0.05) between samples.

3.2.5 Microstructure

Figure 3.6 shows the SEM images of cooked chicken batters without or with various fat substitutes. The SEM images indicated that the addition of different fat substitutes affected the microstructure of cooked chicken batters. A smooth microstructure can effectively bind water, while a coarse microstructure is fragile and has a low water holding capacity [169]. As shown in Figure 3-6, the SEM micrograph of the CK exhibited a granular, rough aspect and large, irregular cavity structure. The cavity in the microstructure of the gel was the water channel [154]. The water channels in the SEM micrograph of the CK had cross throughout the gel network, and it was easy for water to escape from the protein network in macropores, increasing the water loss. In contrast, the water channels in the samples with fat substitutes became thinner or even disappeared; the SEM micrograph exhibited a relatively uniform, compact, homogeneous, and ordered network structure. A possible explanation could be that the cellulose in Ab powder expanded after absorbing water and shrunk the water channel, making the gel network structure more compact. Therefore, it could be inferred that the CY of the samples with various fat substitutes should be greater than that of the CK, which was consistent with the result of CY in our study. In particular, the bridge between adjacent macromolecules was more obvious in the SEM micrograph of the Ab+O sample, followed by Ab and Ab+W samples. A more apparent bridge could prevent the formation of interconnected water channels and assist in generating further independent water cavities, thereby obtaining better gel quality and excellent WHC. This was in agreement with the results of WHC in this study. Previous studies have demonstrated that WHC of meat products is linked to the microstructure [171, 172]. Generally, the diameter of the pores within the protein lattice range from 0.1 to 1.0 μm, which is inversely proportional to the magnitude of the force that immobilizes water [173]. When water is immobilized in pores, the distance of water diffusion to the water/protein interface is shortened than unfixed water, resulting in a more significant proportion of immobilized water molecules to diffuse to the protein/water interface. Therefore, the T2 value of the immobilized water in the pores is lower than that of free water, and as the pore size decreases, the T2 value gradually decreases, approaching that of bound water [172]. Consequently, the addition of three fat substitutes formed more and smaller porous microstructure, which caused a decrease in T2 value, thus resulting in a higher WHC.

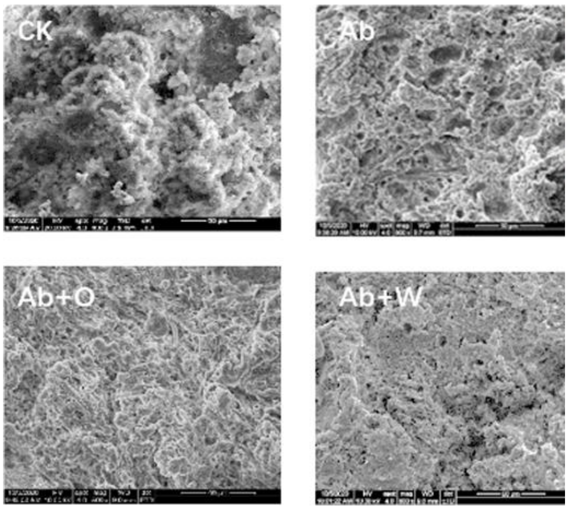

Figure 3.6 – SEM images of chicken gel without or with fat substitutes at 1000 × magnification.

Note: CB: chicken batters; CK: control (100% pork back fat); Ab: CB+80% pork back fat +20% Ab powder; Ab+O: CB + 40% pork back fat + 20% Ab powder+ 40% soybean oil; Ab+W: CB +40% pork back fat + 20% A. bisporus powder + 40% water.

3.3 Effect of the particle size of Ab on the quality of chicken batters

3.3.1 CY, WHC and pH value of chicken batters

As can be seen from Figure3.7, the addition of Ab mushroom powder and soybean oil significantly ($P < 0.05$) improved the cooking yield and water holding capacity of chicken batters, which could be attributed to the abundant cellulose, hemicellulose and lignin in Ab mushrooms [139], which had oil-absorbing and water-binding properties [142, 143]. WHC of D1 was significantly ($P < 0.05$) higher than that of D2, D3 and control group, and there was no significant difference in WHC between D2 and D3. The CY increased significantly ($P < 0.05$) with the increase of the particle size of Ab, which could be attributed to that the larger the size of Ab, the stronger the water absorption capacity[174, 175]. The hydration properties of dietary fiber were affected by physical properties such as particle size and structure, which were one of the important characteristics of dietary fiber [176, 177]. Grinding might influence the hydration properties of dietary fiber [178]. Generally, the smaller the particle size was, the larger the surface area was, and the faster the fiber hydration was [179]. Nevertheless, in some cases, grinding might cause the matrix of the fiber to collapse and reduce the water retention capacity[175].

The pH values of all treatment groups were significantly ($P < 0.05$) higher than that of the control, D1 with the largest particle size had the highest pH value, and D2 and D3 had no significant difference. The possible reason for this was the smaller the particle size was, the easier the acidic components in Ab dissolved out. The higher the pH value, the more conducive to the formation of meat gel, the more refined the gel structure, the greater the gel strength [140]. The result of change trend of pH value was consistent with the change trend of cooking yeild and water holding capacity in present study.

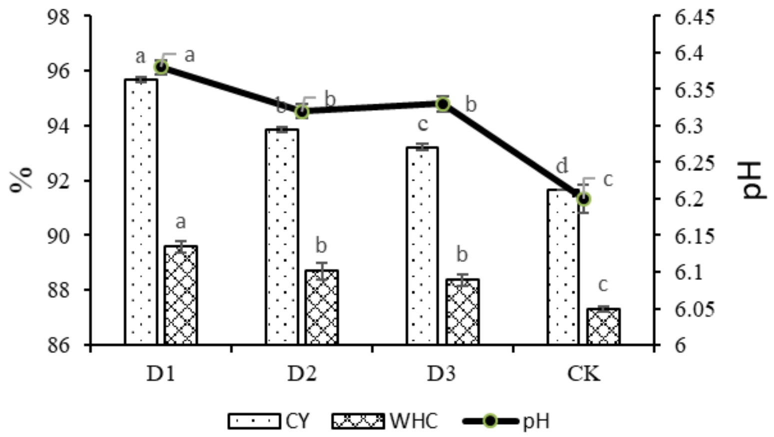

Figure 3.7 – CY, WHC and pH value of chicken batters with different particle sizes Ab.

Note: Values bearing different superscripts are significantly different ($P < 0.05$).

3.3.2 Color of chicken batters

Many factors, such as moisture, fat, myoglobin content, fat and nonmeat ingredients, could affect the color of meat products [180]. In this study, the color change of the treatment group might be attributed to nonmeat ingredient from Ab. As the content of Ab was the same in all treatment groups, the reason of color difference in treatment groups should be due to the different L*, a*, and b* values of Ab powder with different particle sizes.

The L* value of all treatments decreased significantly ($P < 0.05$) compared with the control (Figure3.8). The L* value of chicken gel increased significantly ($P < 0.05$) with the decrease of particle size of Ab, which was due to the fact that the smaller the particle size of Ab, the higher the L* value.

Adding Ab powder significantly ($P < 0.05$) reduced the whiteness value of chicken gel (Figure 3.8). The whiteness value of chicken gel increased with the decrease of particle size of Ab. The whiteness values of each treatment group were different significantly ($P < 0.05$) and all lower than the control significantly ($P < 0.05$), which should attribute to the lower whiteness of Ab itself. Among them, whiteness of the treatment group closest to the blank was the chicken gel with the smallest particle size of Ab, indicating that the smallest particle size had the least impact on the whiteness of the chicken gel.

a* value of D1 (177.93um) was the largest, and there was no significant difference from the control. a* value of D3 (33.73um) was the smallest, and there was a significant difference between the a* values of each treatment group (Figure3.9). The same changes of L* and a* values [181] were found in pork sausages with different particle sizes of celery powders. Fat and vegetable oil were the factors influencing a* value in meat products[182]. In present study, each treatment group had the same amount of fat and soybean oil, so the change of a * values was caused by the significantly ($P < 0.05$) difference of a * values ($P < 0.05$) of different particle size of Ab (Figure3.9).

The b* values of treatment groups increased significantly ($P < 0.05$) compared with the control (Figure3.9). Simultaneously, the b* values of chicken gel increased significantly ($P < 0.05$) as the particle size of Ab increased. The reason might be that Ab with the smaller particle size had larger b* values. Nevertheless, the opposite results were reported in the pork gel with different particle sizes soy okara and the rice meatballs with different sizes of rice bran [183, 184], which may be due to the different color characteristics of raw materials.

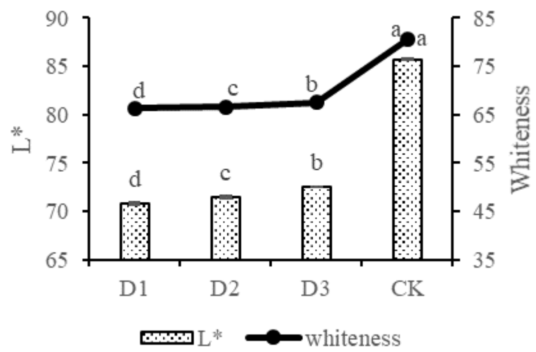

Figure 3.8 – L* value and whiteness of chicken batters with different particle sizes Ab.

Note: Values bearing different superscripts are significantly different ($P < 0.05$).

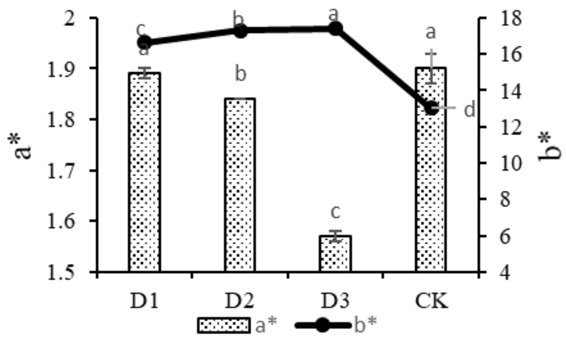

Figure 3.9 – a* value and b* value of chicken batters with different particle sizes Ab.

Note: Values bearing different superscripts are significantly different ($P < 0.05$).

3.3.3 TPA

The particle size of Ab affected the texture characteristics of chicken gel (Figure3.10 and Figure3.11). Including soybean oil and Ab powder with different particle sizes significantly ($P < 0.05$) increased the hardness, springiness and cohesiveness of chicken gel. There was no significant difference in hardness and cohesiveness of samples with different particle sizes of Ab powder and soybean oil. Sánchez-Alonso [185] reported that there was no significant difference in the hardness of samples with different particle sizes of dietary fiber, which was consistent with the results of present study. However, the hardness of pork meatballs increased with the decrease of rice bran particle size [184].

There was no significant difference in the chewiness between all samples and the control group, indicating that the addition of different particle sizes of Ab powder and soybean oil had no significant effect on the chewiness of chicken gels.

The springiness of D2 and D3 samples were significantly ($P < 0.05$) higher than that of D1 samples, indicating that small particle size was more conducive to the springiness of chicken gel. However, soy okara with different particle sizes did not significantly affect the springiness value of pork gel ($P < 0.05$) [183], which was inconsistent with the results of present study and might be due to the different characteristics of raw materials.

Dietary fiber and its particle size could affect the structural characteristics of meat products [184]. Dietary fiber filled the protein network structure as filler, increasing heterogeneity of protein network, thereby enhancing gel strength [165]. Ab powder was rich in dietary fiber [139]. Therefore, Ab could also be used as a filler to affect the texture characteristics of chicken gel network. The different results might be caused by the variety of meat and the type, content and particle size of dietary fiber.

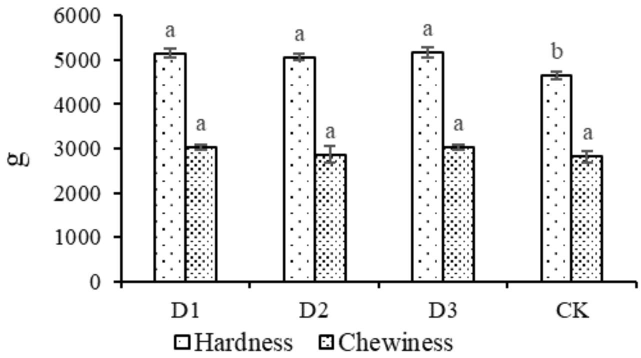

Figure 3.10 – Hardness and chewiness of chicken batters with different particle sizes Ab.

Note: Values bearing different superscripts are significantly different ($P < 0.05$).

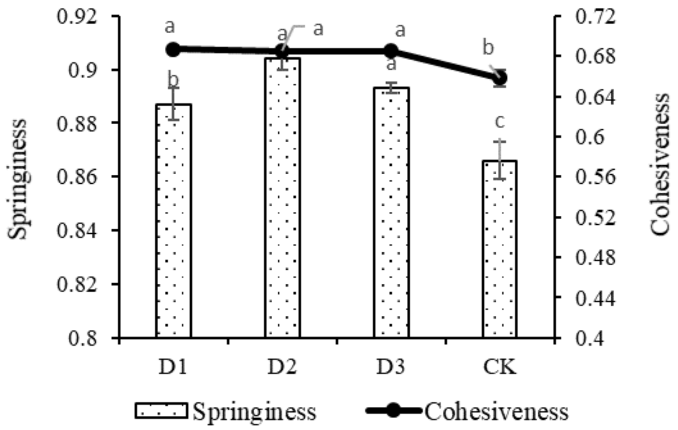

Figure 3.11 – Springiness and cohesiveness of chicken batters with different particle sizes Ab.

Note: Values bearing different superscripts are significantly different ($P < 0.05$).

3.3.4 Correlation analysis

The Pearson correlation coefficients between the particle size of Ab and the quality of chicken batters were shown in Figure 3.12. The particle size of Ab was highly positively correlated with the cooking yield, water holding capacity, a* value and pH value of chicken batters with the Pearson correlation coefficients of 0.973, 0.903, 0.914, 0.821, respectively. The particle size of Ab was highly negatively correlated with the L* value and b* value with the Pearson correlation coefficients of -0.978 and -0.927, respectively. It had a moderately negatively correlation with the elasticity with the correlation coefficient of -0.526 and a weakly correlation with cohesiveness with the correlation coefficient of 0.26. It has no correlation with hardness and chewiness with the correlation coefficients of -0.076 and 0.023, respectively. Therefore, the larger the particle size of Ab, the higher the cooking yield, water holding capacity, a * value and pH value of chicken batters, while the smaller the L * value b * value and springiness. In a word, the particle size of Ab powder should be considered in the production. If the cooking loss and water holding capacity of chicken products are given priority, the large particle size of Ab powder should be selected. If the color and texture characteristics are considered as well, the small particle size of Ab powder should be selected.

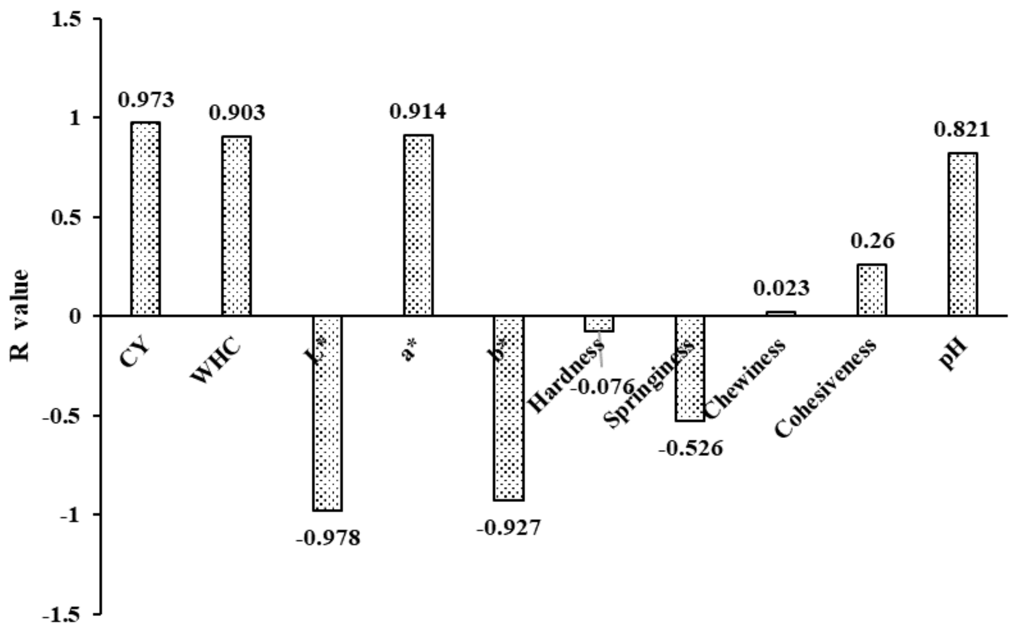

Figure 3.12 – The correlation coefficients between the particle size of Ab and each index

3.3.5 Dynamic rheological properties

The storage modulus (G') and loss modulus (G") reflect important information of muscle protein during the gelation process. The storage modulus (G') reflects the changes in the elastic properties of the gelling network structure and the strength of gel matrix, and the loss modulus (G") reflects the changes in the viscous elements [163].

The thermal gel process of myosin includes two processes: protein denaturation and protein aggregation. When the protein is denatured, the conformation of myosin changes, and the denatured myosin cross-links to form a network structure in the subsequent aggregation process [155]. Figure 3.13 shows that the G' values of all samples decreased at the beginning, which might be due to the fat melting in chicken batters, while the G' values of the three treatment groups were relatively flat compared with the control group, which might be due to the partial replacement of fat in the treatment group. Subsequently, the G' values of all samples first increased, then decreased to the lowest values, and then increase rapidly. The G' values of the control group and the treatment group began to rise at about 43 °C and 35 °C, respectively. At this point, the aggregation of myosin head made the protein denatured, and the network structure began to form slowly [154], which made the G' increase. G' reached its peak when the temperature was between 46°C and 54°C, then G' began to decrease, reaching its peak valley at 62°C. The reason for this might be that the uncoiling of the myosin tail led to an increase in the fluidity of the protein, and destroyed the structure of the gel to a certain extent, thereby reducing G' values [164]. Subsequently, new chemical bonds were formed, and the cross-linking between proteins increased, making G' increase, which was consistent with the change trend of the predecessors on the storage modulus (G') of meat emulsion[162]. Subsequently, as the temperature increased, the G' values of the samples containing Ab were higher than those of the control group, the reason for this might be that the Ab absorbed a large amount of water under the action of high temperature and formed a compact network structure during the heating process, thus making G' increase. The D2 treatment group had the highest G' value, but there was no significant difference from D3. Nevertheless, Wang et al. found that the storage modulus (G') and loss modulus (G″) of pork mince decreased with the decrease of soybean residue particle size, which was inconsistent with the results of present study. This might be due to the premixing of Ab granules with soybean oil in present study. The smaller particles of Ab can fully disperse the soybean oil upon premixing, which reduced the accumulation of oil droplets to a certain extent. The protein content used for emulsification reduced, and more protein was used for coagulation and the formation of the gel, thereby the chicken gel had a better network structure.

The increase in the viscosity of chicken batters was related to the water/fat binding capacity and the stability of the emulsion [25]. The final G" values of all treatment groups were higher than that of the control (Figure 3-14), indicating that the combination of Ab and soybean oil with different granularities improved the emulsion's stability. The change trend of loss modulus G" was similar to that of storage modulus G'. The value of G " first increased between 33 °C and 36 °C, reached the peak between 50°C and 54°C, then dropped to the bottom at 63 °C, and then increased rapidly and reached the maximum at 80 ° C (Figure 3.14). Among them, the value of G" in the gel of D2 treatment group was the highest, but there was no significant ($P < 0.05$) difference from that of D3. The G" values of chicken batters were lower than G' value during the entire heating process of gel formation, indicating that elasticity was more dominant than viscosity in chicken batters system.

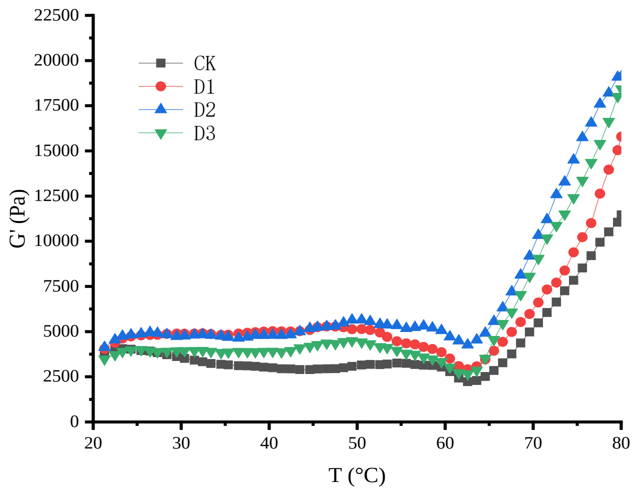

Figure 3.13 – Effect of temperature on the storage modulus (G') of chicken batters with different particle sizes of Ab

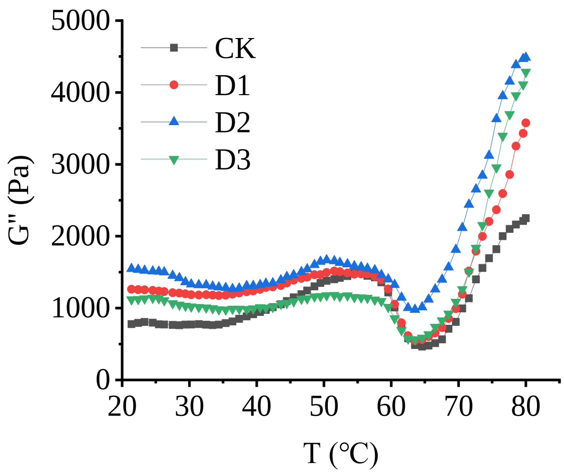

Figure 3.14 – Effect of temperature on the loss modulus (G") of chicken batters with different

particle sizes of Ab

3.3.6 LF-NMR spin–spin relaxation (T2)

LF-NMR technology can illustrate the state of water in the gel system. The continuous distribution of T2 values are presented in Figure 3.15, Figure 3.16, Figure 3.17, and Figure 3.18. T2 relaxation time and corresponding peak areas percentages of water from chicken gel are shown in Table 3.6.

Water in the muscle structure can be classified into three types according to the activity. The first type of water, T2b, tightly bounding to myofibrillar protein, has a relaxation time of between 0 and 10ms. The immobilized water, T21, locating in the myofibrillar protein matrix, has a relaxation time of between 10 and100ms. The free water, T22, locating outside the myofibrils , is the most fluid water component in the meat and has a relaxation time of 100 and 400 ms [168, 186]. The lower the proton mobility in the gel, the faster the T2 relaxation time, and the more the T21 peak is to the left[167].

It can be seen from Table 3.6 that the incorporation of Ab and soybean oil caused the T21 shifted toward faster relaxation times compared with the control group, and D1 had the fastest T21 ($P < 0.05$). This may be due to the fact that the active groups of dietary fiber in Ab enhanced the interaction between proteins, and then promoted the self-binding of proteins, forming a firm network, thus limiting the mobility of water protons [171]. In addition, Ab filled in inter-gel and the hydrophilic groups of dietary fiber in Ab combined more hydrogen proton and the hydration of gel network is faster [34]. Compared with the control, the addition of Ab significantly increased the P2b of chicken gel ($P < 0.05$) and accompanied by a remarkable decrease in PT22 ($P < 0.05$), which indicated that adding of Ab and soybean oil promoted the tight bounding of water and protein, reduced the proportion of free water, and improved water retention.

Among the treatments, D1 had the largest PT2b and the smallest PT21, while PT22 had no significant difference with D2 and D3, which might be attributed to the physical retention of water by the porous surface of the dietary fiber in Ab mushroom. Meanwhile, it indicated that the water in D1 flowed to the bound water direction, which significantly ($P < 0.05$) increased the stability of water and made D1 had the largest water holding capacity, which was consistent with the results of CY and WHC in present study. These results suggested that it was necessary to optimize the particle size of mushroom powder for the gel system.

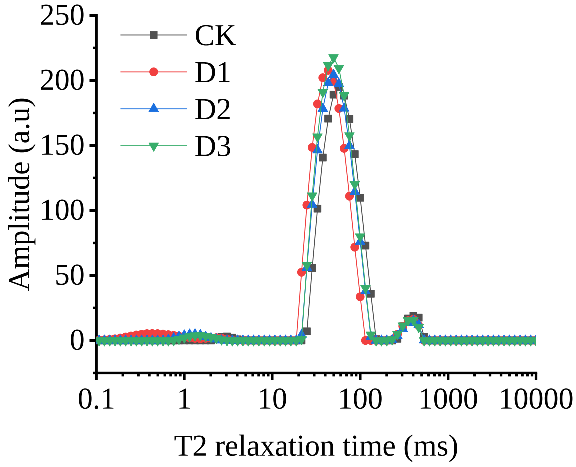

Figure 3.15 – T2 relaxation time of chicken gel with different particle sizes of Ab

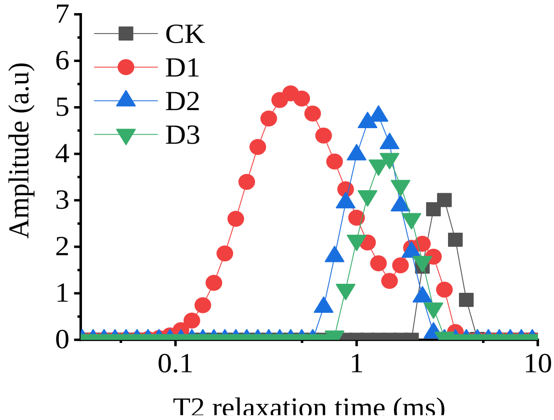

Figure 3.16 – T2b relaxation time of chicken gel with different particle sizes of Ab

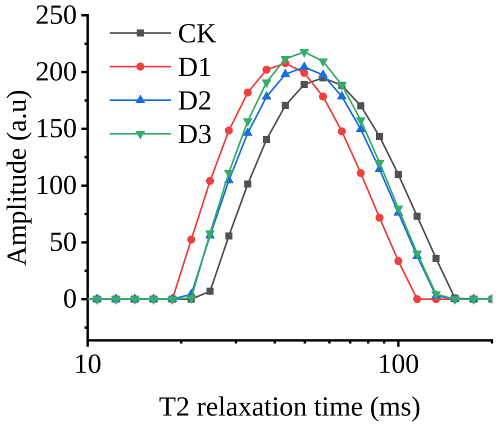

Figure 3.17 – T21 relaxation time of chicken gel with different particle sizes of Ab

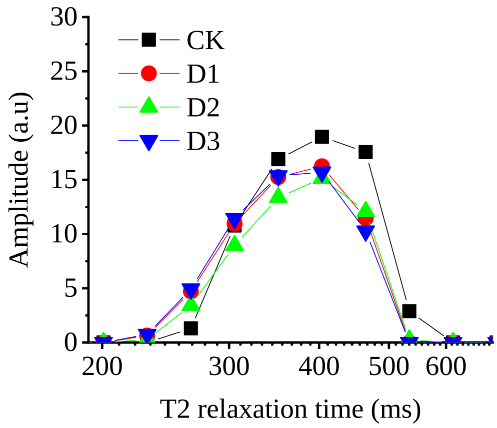

Figure 3.18 – T22 relaxation time of chicken gel with different particle sizes of Ab

Table 3.6 – T2 relaxation times and corresponding peak areas percentages of water from chicken gel with different particle sizes of Ab

Treatments	Traits					
	T2b	T21	T22	PT2b(%)	PT21(%)	PT22(%)
CK	2.01±0.04a	21.54±2.15a	231.01±13.62a	0.63±0.21d	95.25±1.34a	4.12±1.87a
D1	0.03±0.01c	18.74±2.53b	200.92±13.42b	3.83±0.24a	92.70±1.42b	3.46±0.31b
D2	0.57±0.08b	18.74±2.47b	200.92±12.38b	1.67±0.22b	95.13±1.67a	3.20±0.37b
D3	0.66±0.10b	18.74±3.01b	200.92±12.98b	1.21±0.32c	95.48±1.51a	3.30±0.45b

Note: a-d Different letters in the same column indicate significant differences ($P < 0.05$).

3.3.7 Microstructure of cooked chicken batters

Figure 3.19 displays SEM graphs of chicken gels added without and with different particle sizes of Ab and soybean oil. It can be seen from Figure 3.19 that the microstructure of chicken gel network changed obviously with the decrease of the particle sizes.

The SEM image reflected the microstructure of the chicken gel and can be used for explaining the water holding capacity of the gel. Smooth gels could effectively bind water, while coarse gels were fragile and had low water holding capacity[169].The control group had rough surface and large, irregular cavity structure, whereas the treatments showed uniform, compact, homogeneous and ordered network structure. The pore in the microstructure of gel was water channe [154], it was easy for water to escape from the protein network in macropores, resulting in the increase of water loss. Therefore, it can be inferred that the cooking loss of all treatment groups should be less than that of the control, which was consistent with the result of WHC in present experiment.

Among the SEM images of D1, D2 and D3, D1 showed a larger pore size and rougher surface, the possible reason was that the Ab with larger particles had greater water holding capacity and swelling characteristics than the small Ab. Most of the water in the gel was captured by Ab powder by hydrogen bonding, then the mushroom powder swelled and filled the gel network gap of chicken protein, creating big cavity. Nevertheless, Ab with small particle size had less water holding, oil holding and swelling capacity. As filled into the protein network structure, Ab with small particle size and soybean oil made the gel network structure more uniform. Nevertheless, the SEM images of D2 and D3 did not show a distinct difference, the possible reason was the absorbable and expanding Ab powder just filled the gaps in the protein three-dimensional network structure, showing a more uniform microstructure.

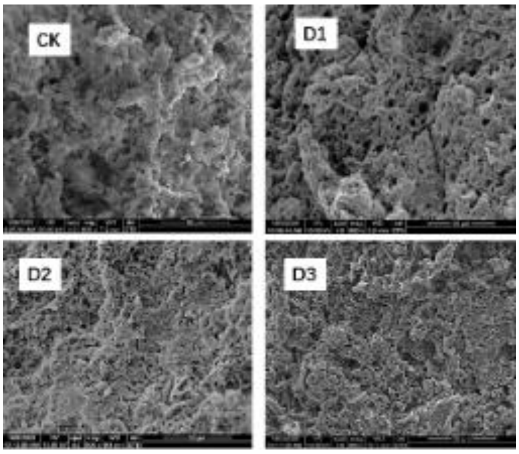

Figure 3.19 – Typical scanning electron micrographs (SEM) at 2,000× magnification of chicken batters with different particle sizes of Ab.

3.4 The effect of Ab on the gel properties of chicken myofibrillar protein

3.4.1 Gel strength and WHC of MP gels

The gel strength indicates the aggregation ability of the protein. The stronger the gel strength, the tighter the gel structure, and the more stable the gel formed. Higher WHC means stronger protein binding capacity to water. Dense and uniform gel structures typically possess high WHC and low cooking losses, resulting in improved meat product yields. Therefore, gel strength and WHC are vital parameters for evaluating the gelation property of MP composite gels [155]. As seen in Figure3.20, an increase in mushroom powder levels enhanced gel strength and WHC. In particular, compared to those of the CK, gel strength and WHC of the sample with 6% mushroom powder (T6) increased by

100.1% and 37.6%, respectively, which could be attributed to the rich protein and dietary fibre in mushrooms [139]. On the one hand, the incorporation of protein from mushrooms increases the total protein content in the system, resulting in higher aggregation ability of the protein and gel strength. On the other hand, the fibre from mushrooms possesses good water-binding properties, thus improving the WHC of MP gels [142]. It has been reported that adding insoluble sugarcane dietary fibre (SIDF) (0.5% to 2%) significantly increased the gel strength and WHC of MP gels ($P < 0.05$) [154]. Another important reason for the improvement in WHC of MP gels may be that fibres in mushrooms interact with proteins during heating, forming dense network structures, retaining moisture in the thermal gels, and consequently enhancing WHC [187].

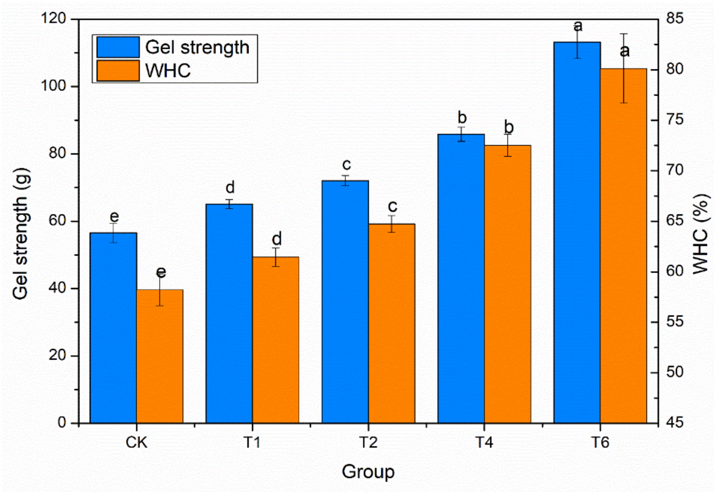

Figure 3.20 – Gel strength and WHC of MP gels with various levels of Ab mushroom.

Note: Different letters in the same column mean significant differences ($P < 0.05$). CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms (w/w), respectively.

3.4.2. TPA of MP gels

The texture is a major factor in the acceptability and quality of food products[136]. As shown in Table 3.7, with the addition of Ab, the hardness gradually increased, while the CK exhibited the minimum hardness ($P < 0.05$). These results agreed with our previous discovery, wherein Ab powder increased the firmness of low-fat chicken batter [188, 189], which might be related to the rich dietary fibre contained in Ab mushrooms filling the gaps of the gel network matrix, thereby improving the gel strength. Thus, when the amount of mushroom added increased, more dietary fibre packed into the gel structure, making it denser and more compact, thus increasing the firmness. Moreover, Ab significantly enhanced the springiness of the MP gels compared with CK ($P < 0.05$), and the springiness increased with increasing concentrations of Ab (Table 3.7), which might also be due to the rich dietary fibre contained in Ab mushrooms [139]. Many studies have indicated that dietary fibre can improve the hardness and springiness of MP gels [155, 190-192]. The gumminess of the MP composite gel containing Ab mushroom showed the same increasing trend with an increasing level of Ab ($P < 0.05$), which might be due to the strong viscous characteristic of dietary fibre in Ab. The present results accord with the previous finding [155] that regenerated cellulose fibres significantly strengthened the gumminess of MP gels (P < 0.05). The cohesiveness of the MP gel was considerably higher than that of the CK when the Ab level exceeded 1% (T1) ($P < 0.05$) (Table 3.7). However, the cohesiveness decreased significantly ($P < 0.05$) above 4 % (T4). These results demonstrate that the voids in the MP gel network can only accommodate a specific amount of Ab. The excess Ab may interfere with protein aggregation and cross-linking, making protein gels less cohesive, which may relate to the ash contained in the Ab. The results of this study are consistent with our previous findings that a particular amount of Ab as a fat replacer can indeed enhance the texture of the chicken batter[188]. Generally, the rheological properties of MP determine the viscoelastic properties of their heat-induced gels. Therefore, studies on dynamic rheology are of great significance to further explore the effect of Ab on MP gelation.

Table 3-7 The textural properties of MP gels with various levels of Ab mushroom

Treatments	Hardness(g)	Springiness	Cohesiveness	Gumminess(g)
CK	61.59±4.22e	0.572±0.014e	0.660±0.011c	21.94±3.43e
T1	75.63±5.06d	0.741±0.035d	0.677±0.007bc	29.09±3.70d
T2	99.48±7.38c	0.814±0.022c	0.691±0.028ab	43.12±6.40c
T4	147.53±12.34b	0.872±0.011b	0.740±0.023a	102.99±19.30b
T6	178.24±16.77a	0.934±0.041a	0.685±0.015b	214.91±28.57a

Note: Different letters in the same column mean significant differences ($P < 0.05$).CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms (w/w), respectively.

3.4.3. Dynamic rheological properties

The storage modulus (G′) (Figure 3.21A) and loss modulus (G″) (Figure 3.21B) show the viscoelasticity of MP composite solutions with different levels of Ab powder. G′ and G″ reflect the continuous changes of MP interactions to some extent [193]. With a temperature rise, the G' value of each sample showed a similar trend. It rose initially and peaked at about 56°C, then decreased and hits bottom at about 61°C. It then again rapidly increased and plateaued at 80°C (Figure 3-21A). Generally, at the initial stage of temperature rise, the head of myosin binding to each other produced a weak gel network structure, and G' increased [194]. The reduction in G' (56 °C–61 °C) was related to network destruction and increased protein mobility induced by the unfolding of the myosin tail [164]. Finally, the G' increased, suggesting that the cross-linking and aggregation of proteins formed a stable and elastic gel with a more robust network structure [169]. These results accord with the typical features of MP gelation [155]. The G′ values of MP composite solutions with various amounts of Ab powder were higher than that of the CK during heating. It can be inferred that Ab mushroom might enhance the covalent crosslinking in MP. Thus, adding Ab mushroom to the MP can quickly strengthen G' throughout the heating process (Figure 3-21A). This result may partly be explained by the presence of insoluble components from the Ab mushrooms, such as cellulose and lignin, which enter the voids of the protein matrix, resulting in a denser gel structure. Furthermore, the protein and polysaccharides from Ab mushrooms can form a net-like structure, strengthening the MP-Ab gel structure [139].

The G" values of MP containing different amounts of Ab mushrooms showed a similar trend, compared with G' values (Figure 3-21B). The G" value represents the viscosity properties of the gel network structure. The Ab-added MP hybrid solutions exhibited higher G" values compared with the control during heating. Meanwhile, G" increased with an increase in the level of Ab. It shows that the incorporation of Ab enhances the viscosity of the gel system. It has been reported [155] that a higher viscosity of the protein gel is related to its better water-binding capacity. The results of G" and WHC in the present study are consistent. Therefore, Ab has good water retention capacity, which benefits the viscosity of the MP gel.

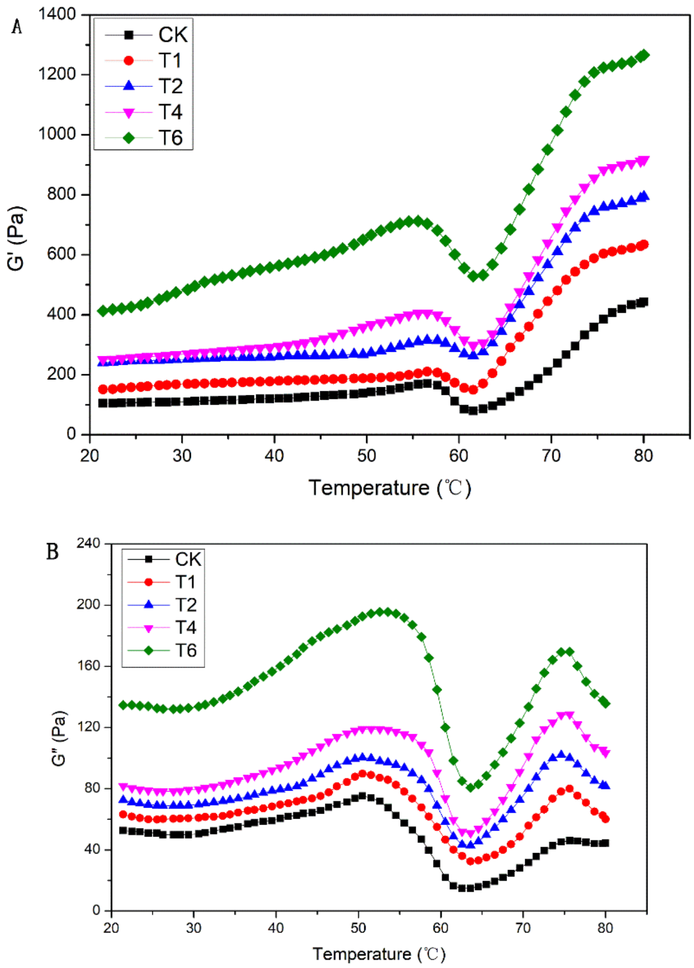

Figure 3.21 – The storage modulus G' (A) and loss modulus G'' (B) of MP gels with various levels of Ab mushroom.

Note: CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms (w/w), respectively.

3.4.4 LF-NMR spin-spin relaxation (T2)

In the gel system, the existing state of water molecules can be estimated by T2 relaxation time, which can be measured by LF-NMR [195]. Therefore, in this study, the existing state of water molecules in the MP-Ab gel system was checked by T2.

Figure 3.22 shows a representative distribution of NMR T2 measurements. As shown in Figure 3.22, three separate peaks, T2b, T21, and T22, appeared, capturing 1-3%, 87-94% and 4-11% of total water, respectively. The T2 are 2-4 ms, 75-100 ms, and 1072-1630 ms, respectively, with a total relaxation time from 0.01 to 10,000 ms. In general, the first peak, T2b, represented bound water, which was bonded to MP by strong H-bonds [167]. The second peak, T21, corresponded to immobilized water. This part of the water was tightly connected with the monolayer water molecules and trapped in the MP matrix [167]. The third peak, T22, was attributed to free water, located outside MP, which had maximum fluidity in three types of water [162]. Table 2 displays three T2 (T2b, T21, and T22) and corresponding peak area fractions (PT2b, PT21, and PT22) of water in MP gel. As shown in Table 3-8, the Ab-added MP composite gel exhibited a shorter relaxation time than the control. T2b and T21 did not change with the addition of Ab mushroom. At the same time, T22 decreased significantly ($P < 0.05$), indicating that the incorporation of Ab increased the binding state of water and protein in MP gel and reduced the degree of freedom of water. These results could be attributed to the active groups of dietary fibre from Ab, which reinforced the interaction between proteins, forming a firmer network. As a result, the mobility of water protons was restricted, and the degree of freedom of water was weakened [181]. In addition, Ab filled the inter-gel spaces, and the hydrophilic groups of dietary fibre from Ab bound more hydrogen protons, resulting in a faster hydration process for the gel matrix [34]. P21 showed an upward trend, while P22 showed a downward trend with the addition of Ab mushroom (Table 3.8), which indicated that Ab could convert free water to immobilized water. This conversion is generally considered to result in an improvement in the WHC [154]. The results of P21 and P22 agree with the WHC values obtained in the present study.

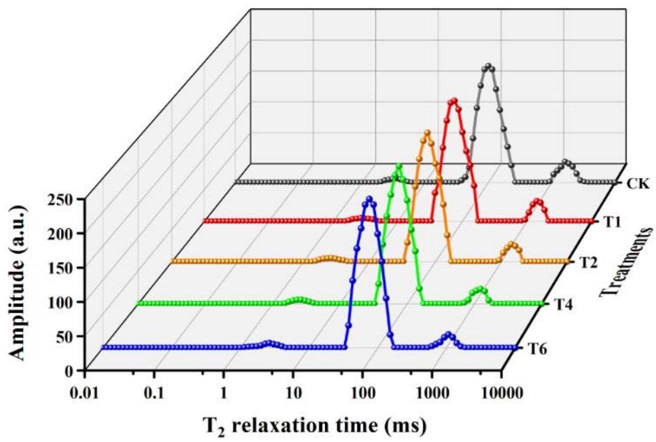

Figure 3.22 – The continuous distribution of T2 relaxation time of MP gels with various levels of Ab mushroom.

Note: CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms (w/w), respectively.

Table 3.8 – T2 relaxation times and corresponding peak areas percentages of MP gels with various levels of Ab mushroom

Treatments	T2b (ms)	T21 (ms)	T22 (ms)	PT2b (%)	PT21 (%)	PT22 (%)
CK	3.51±0.28a	100±13.03a	1629.75±125.72a	2.00±0.66a	87.32±0.36d	10.68±0.19a
T1	2.66±0.22a	75.65±12.14b	1417.47±122.46b	1.90±0.43a	89.80±0.43c	8.29±0.36b
T2	2.66±0.25a	75.65±13.25b	1417.47±120.58b	2.07±0.15a	90.66±0.45c	7.27±0.15c
T4	2.66±0.31a	75.65±12.06b	1232.85±60.61c	2.06±0.37a	91.80±033b	6.14±0.24d
T6	2.66±0.28a	75.65±12.34b	1072.27±95.59d	2.12±0.25a	93.13±0.51a	4.75±0.18e

Note: Different letters in the same column mean significant differences ($P < 0.05$).CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms, respectively.

3.4.5 Microstructure of MP gels

The SEM graphs (Figure 3.23) indicated that the denaturation and aggregation of MP produced a gel structure with an irregular appearance. The microstructure of MP gel containing different Ab mushroom concentrations exhibited other pronounced appearances. The micrograph of the CK demonstrated a rougher aspect, discontinuous, disordered gel matrix, and large, irregular cavity structure. The incorporation of Ab mushroom increased the density of MP gel structure, leading to a more uniform, compact, homogeneous, and ordered gel microstructure than the CK. Moreover, the cavity appeared thinner or even disappeared with an increase in Ab mushroom concentration (Figure 3.23). It is reported that the WHC of meat products was related to the gel microstructure [193]. These results of the microstructures are consistent with the WHC results, demonstrating that Ab mushroom promotes the improvement of MP gel structure. One possible explanation for this is that the abundant cellulose in the Ab powder may have absorbed water and swelled, thereby reducing the water channels through the filling effect, forming a gel network structure by itself. These beneficial properties of Ab mushroom can make the protein gel denser, which helps to retain moisture and increase the gel hardness. However, when the content of Ab mushroom was 6%, a sheet structure appeared due to its excessive density, which resulted in a decreased cohesiveness compared with the gel containing 4% Ab powder. The reason for this may be that the addition of excessive Ab mushroom in the external phase could have been sufficient for it to self-aggregate, which is adverse for the gel structure [155].

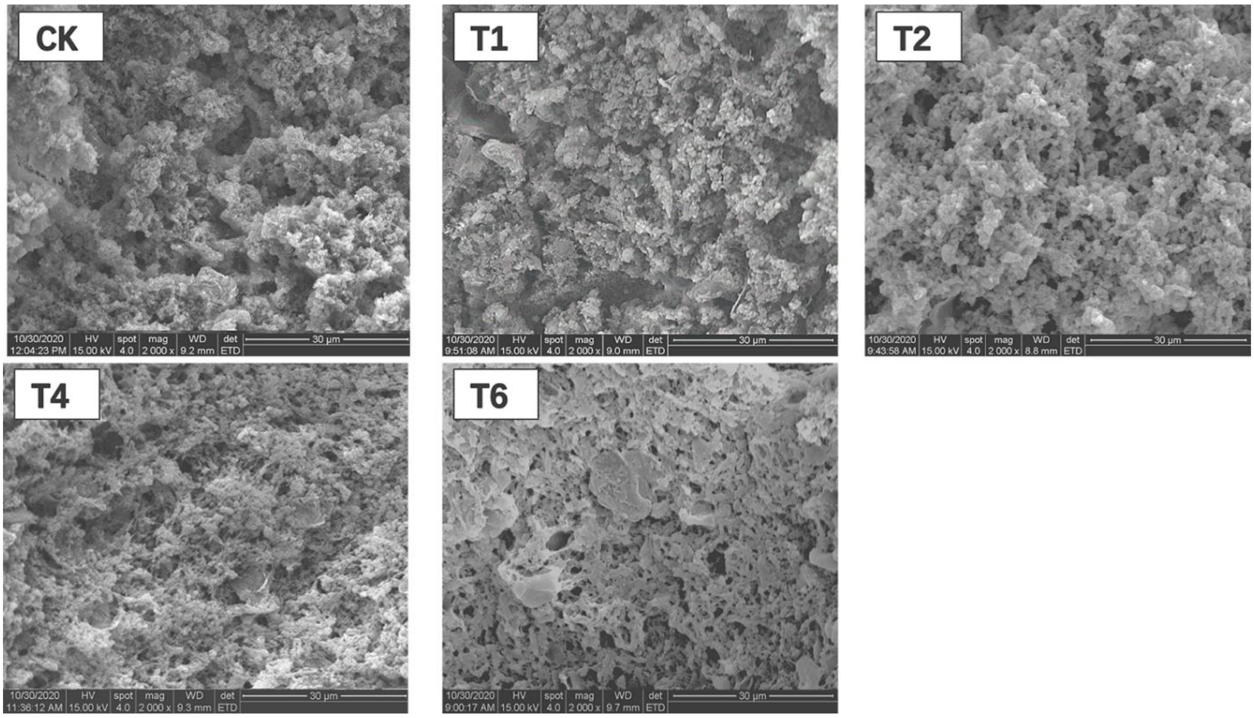

Figure 3.23 – SEM images of MP gels with various levels of Ab mushroom.

Note: CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms (w/w), respectively.

3.4.6 Protein secondary structure

To elucidate the mechanism of the effect of Ab mushroom on gelation, the protein secondary structure was assessed using FTIR spectroscopy (Table 3.9). Compared with the CK, the α-helix content of the Ab-added MP complex gel was significantly reduced ($P < 0.05$). Nevertheless, the percentages of the β-sheets, β-turns, and random coil significantly increased ($P < 0.05$). The previous study has shown that a decrease in α-helix and an increase in β-sheet is beneficial for protein gelation [155]. As previous report [196, 197], the transformation between α-helix and β-sheet often occurs in the protein secondary structure. α- helix is one of the most common protein secondary structures, which is stabilized mainly by hydrogen bonds between -CO and NH- of a polypeptide chain. Therefore, it can be inferred from the present findings that the reason for the increase of the β-sheet structure may be that the Ab mushroom breaks the hydrogen bond of the α-helix after entering the MP gel structure, promoting the unfolding of the α-helix. These results agree with the previous report that the WHC and G' values of MP gels were significantly improved with the conversion of α-helices to β-sheets and β-turns [155]. Therefore, the above results indicate that the incorporation of Ab could alter the secondary structure of chicken MP to enhance the WHC and viscoelasticity of Ab-MP gels.

Table 3.9 – The secondary structure fractions of MP gels with various levels of Ab mushroom

Treatments	α-Helix	β-Sheet	β-Turn	Random coil
CK	34.25±2.34a	31.51±2.05d	23.20±0.19d	11.04±0.08d
T1	30.22±1.34b	34.85±1.23c	23.63±0.22c	11.30±0.06c
T2	26.97±2.15bc	36.85±1.61bc	24.68±0.35bc	11.50±0.16bc
T4	23.01±1.16c	39.25±1.02b	25.95±0.18b	11.79±0.14b
T6	20.25±1.35d	41.17±0.87a	26.59±0.24a	11.99±0.09a

Note: Different letters in the same column mean significant differences ($P < 0.05$). CK, T1, T2, T4, and T6 represent treatments containing 0%, 1%, 2%, 4%, and 6% Ab mushrooms, respectively.

CONCLUSION OF SECTION 3

1. The pH, emulsion stability, texture, color and microstructure of chicken batters (containing 20% pork back fat) were systematically studied by using Ab powder to replace 2, 4, 6, 8, 10, 20, 30, 40, and 50% pork back fat respectively. The results showed that the pH value and cooking yield of chicken batters improved significantly when the fat replacement percentage was greater than 2%. The hardness, springiness, cohesiveness, and chewiness of chicken batters increased significantly when the fat replacement percentage was 20%- 50%, 8%-20%, 6%-8%, and 20%-50%, respectively. Taken together, when Ab powder replaced 20% of the fat, the texture properties of the chicken batters were significantly improved, and the microstructure was more compact and uniform. However, the replacement of fat with Ab significantly changed the color of chicken batters, resulting in a significant decrease in the L* and whiteness value and a significant increase in a* and b* value of the chicken batters.

2. The gel properties, rheological properties, water distribution and microstructure changes of chicken batters (including 20% pork back fat) were systematically studied when Ab was used alone to replace 20% fat, Ab was mixed with water or soybean oil in 1:2 to replace 60% pork back fat. The result showed that the addition of Ab powder or the combination of Ab powder and soybean oil or water transforms part of free water into immobilized water and significantly ($P < 0.05$) improves CY, WHC, texture, and

rheological properties of the chicken batter. However, the color of the chicken batter changed significantly ($P < 0.05$). Particularly, the chicken batter with Ab powder and soybean oil showed a higher CY, WHC and texture parameters, a more uniform and dense microstructure, and a relatively minor color change. Therefore, the results suggested that a combination of Ab powder and soybean oil could be utilized to replace 60% pork back fat for the production of reduced-fat and healthy chicken products.

3. The changes of cooking yield, water holding capacity, pH, color, rheology, water distribution and microstructure of chicken batters (including 20% pork back fat) were systematically studied when Ab powder with different particle sizes and soybean oil were mixed in 1:2 instead of 60% pork back fat. The results showed that the bigger particle sizes of Ab increased the cooking yield, water holding capacity, and pH value of chicken batters. With the reduction of particle sizes of Ab powder, the L* values, whiteness, and b* values of chicken batters increased significantly ($P < 0.05$). The decrease of the particle size of Ab could improve the springiness. The particle size of Ab was highly correlated with the cooking yield, water holding capacity, L*, a*, b*value and pH value of chicken batters. It had a moderate correlation with springiness. The combination of large particle sizes of mushroom powder and soybean oil increased the water holding capacity of gel by converting the free water to the bound water in the chicken batters system. The combination of small particle sizes mushroom powder and soybean oil improved the viscoelasticity of the chicken batters and made the microstructure more compact and uniform.

4. The effects of Ab powder on gel strength, water holding capacity (WHC), texture, rheological behaviour, LF-NMR spin–spin relaxation (T2), microstructure and protein secondary structure of the MP gel system were systematically studied. The results indicated that the gel strength, WHC, G’ value and G" value were significantly improved when the addition of Ab powder increased from 0% to 6% ($P < 0.05$). Meanwhile, the T2 relaxation time shortened, and free water transformed into immobilised water. The texture of the gel improved when 1%–4% Ab powder was added compared to the control. Furthermore, Ab filled in the gel network and promoted the unfolding of MP a-helix and the formation of MP b-sheet during the thermal denaturation of MP, leading to a dense

aggregated network structure. These results suggest that Ab mushroom powder can enhance the gel properties of chicken myofibrillar proteins.

SECTION 4 THE RESEARCH OF THE RECIPE COMPOSITION, QUALITY PROPERTIES, AND TECHNOLOGICAL SCHEMES OF THE SAUSAGES

4.1 Optimization of the recipe composition of the sausage

4.1.1 Optimization of starch addition in sausage

Figure 4.1 shows the effect of different amounts of potato starch on the taste, color, flavor, texture and general acceptability of chicken sausage. It can be seen that the addition of potato starch had the greatest impact on the general acceptability, texture and taste of sausage, followed by flavor and color. Among them, the sausage containing 5% potato starch scored higher than the other groups in general acceptability, taste, color and flavor, and its texture score also reached an acceptable level of 5.8. Therefore, the optimal addition amount of potato starch was 5%.

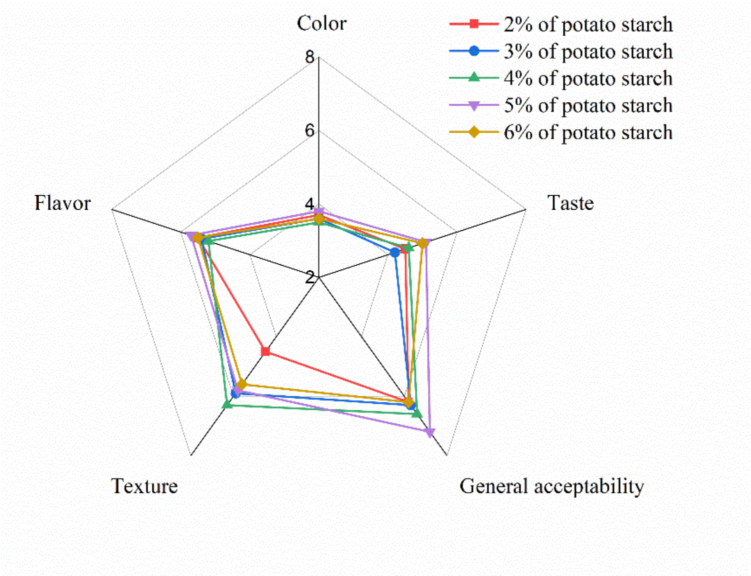

Figure 4.1 – Effects of potato starch addition on sensory scores of chicken sausages

4.1.2 Optimization of chicken essence addition in sausage

Figure 4.2 shows the effect of different chicken essence additions on the taste, color, flavor, texture and general acceptability of chicken sausage. It can be seen that after adding chicken essence, the five sensory properties of chicken sausage were all improved. Among them, the addition of 0.1% chicken essence scored the highest in terms of general acceptability, taste, color, flavor and texture, and its flavor, texture and general acceptability all reached more than 6 points, reaching the level of liking. Its color and taste were both above 5 points, which were acceptable, so the optimal addition amount of chicken powder was 0.1%.

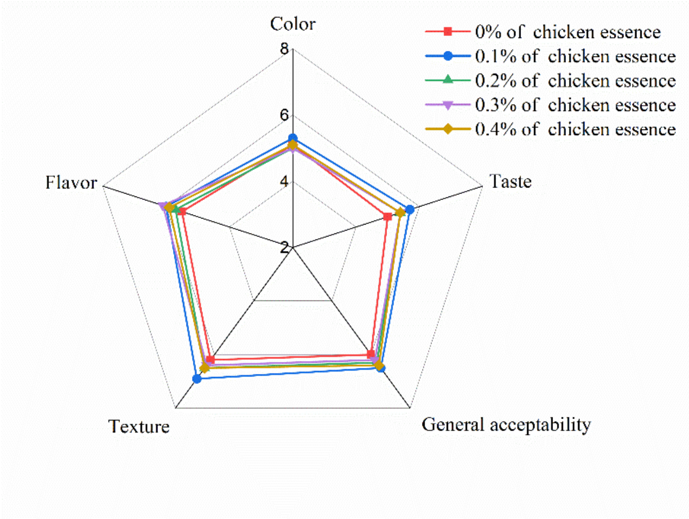

Figure 4.2 – Effects of chicken essence addition on sensory scores of chicken sausages

4.1.3 Optimization of refined salt addition in sausage

Figures 4.3 shows the effect of different refined salt additions on the taste, color, flavor, texture, and general acceptability of chicken sausages. It can be seen that changes in the amount of salt added had a greater impact on the texture, flavor and general acceptability of the sausage. The sausages with 1.4% salt had the highest scores for taste, color, flavor, texture, and general acceptability. Its flavor, texture and general acceptability all achieved more than 7 points, reaching the level of liking. The color and smell were both 5.6 points, which were close to the level of liking. Therefore, the optimal amount of refined salt added was 1.4%.

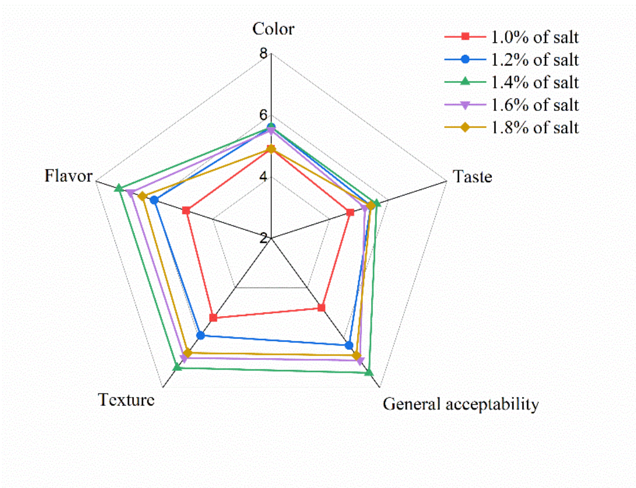

Figure 4.3 – Effects of refined salt addition on sensory scores of chicken sausages

4.1.4 Optimization of Ab and soybean oil addition in sausage

Figure 4.4 shows the sensory scores of chicken sausages with different Ab and soybean oil addition. It can be seen from the figure 4.4 that the incorporation of Ab and soybean oil increased consumers' preference for the flavor, texture, taste and general acceptability. However, the three groups of chicken sausages containing Ab and soybean oil all scored higher than 5 points in the color item, indicating that consumers could accept the change in the color of the sausage caused by the addition of Ab and soybean oil. Among them, the flavor, texture and general acceptability of the T60 group all reached more than 7 points, which were favored by consumers, and the taste and color of T60 were also 5.6 points, indicating that consumers were acceptable. Taken together, the T60 group scored higher, and consumers were more willing to accept the low-fat chicken sausages that replace 60% of the fat with Ab and soybean oil.

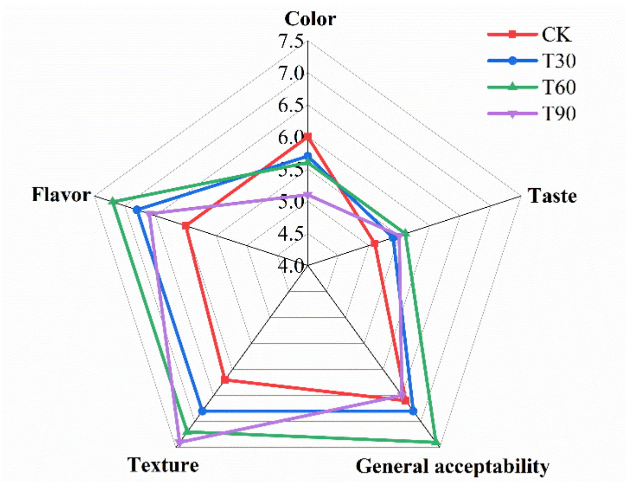

Figure 4.4 – Sensory score of the chicken sausages with different Ab and soybean oil addition

4.2 The research of the quality properties of the sausage

4.2.1 Properties of Ab powder and chemical composition of the chicken sausage

It can be seen from Table 4.1 and Table 4.2 that the ash and protein content in the Ab powder was 10.15% and 22.14% respectively, while the fat content was only 2.53% and the carbohydrate content was as high as 58.22%. As expected, the content of fat in the sausage decreased significantly, and the ash and carbohydrate content increased significantly with increasing fat replacement ($P < 0.05$), the reason for this should be that

Ab powder was rich in ash and carbohydrates and low in fat (Table 4.1). However, the protein content of the T90 groups decreased significantly compared with the CK group (Table 4.2). This may be due to the fact that two thirds of the Ab premix is soybean oil, which contained no protein, while pork back fat contains a certain amount of protein. Compared with the control group, there was no significant change in protein content of T30 and T60 due to the small amount of fat replacement. In this study, there was no significant difference in the moisture content of all the treatments, the reason for this may be that there was no significant difference in the cooking yield among the four treatments (Figure 4.1). The carbohydrate content of T60 and T90 was significantly higher than that of CK and T30 ($P < 0.05$), and the carbohydrate content increased significantly with the increase of fat replacement. The reason for this should be that Ab powder were rich in carbohydrates (Table 4.1). Replacing the fat with Ab and soybean oil significantly reduced the energy value of the sausage, but there was no significant difference between the energy values of T30, T60 and T90.

Table 4.1 – The properties of Ab powder

Moisture/%	Ash/%	Protein/%	Fat/%	Carbohydrate/%
6.96±0.11	10.15±0.34	22.14±0.53	2.53±0.05	58.22±0.73
L*	a*	b*	PH	
51.26±0.49	1.91±0.09	14.7±0.13	6.53±0.05	
Essential aminal acid (g/100g total aminal acid)				
Val	Met	Ile	Leu	Phe
1.19±0.10	0.27±0.01	1.02±0.03	1.44±0.09	0.97±0.02
His	Lys	Thr	Arg	
0.41±0.01	1.21±0.10	1.03±0.08	0.98±0.02	
Non-essential aminal acid (g/100g total aminal acid)				
ASP	Glu	Ser	Gly	Pro
1.89±0.11	5.02±0.24	0.83±0.02	1.11±0.08	1.01±0.05
Ala	Cyst	Tyr		
1.79±0.02	0.06±0.01	0.77±0.02		

Table 4.2 – Proximate composition of the chicken sausage with Ab powder and soybean oil

Items	CK	T30	T60	T90
Moisture (g/100g)	61.06±1.32a	64.58±2.22a	61.85±1.18a	62.71±1.25a
Ash (g/100g)	0.73±0.07c	0.76±0.05c	1.54±0.01b	1.92±0.06a
Protein (g/100g)	15.38±1.40a	14.36±0.99a	12.51±1.48ab	11.9±0.94b
Fat (g/100g)	18.56±0.31a	16.74±0.24b	14.8±0.34c	12.9±0.21d
Carbohydrate (g/100g)	4.27±0.37c	3.56±0.35c	9.30±0.34b	10.57±0.35a
Energy (kcal/100 g)	245.64±9.87a	222.34±7.52b	220.44±10.34b	205.98±7.05b

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$).

4.2.2 Amino acids profile

As can be seen from Table 4.3 that the contents of the essential amino acids and EAA/NEAA of the T30 sample were significantly increased ($P < 0.05$) and the content of non-essential amino acids in the T90 sample was significantly increased compared to the CK. Many studies have reported the improvement of the protein composition in meat products by adding non-meat protein ingredients, such as seaweed, pulses, chlorella and spirulina [198, 199]. In the present case, the addition of Ab powder and soybean oil did not significantly change the amino acid profile, except for T30, and the ratio between essential and non-essential amino acids remained at 0.98-1.06. The main amino acids in sausage were leucine (1.10–1.32 g/100 g), lysine (1.18–1.37 g/100 g), aspartic acid (1.23–1.38 g/100 g), gluten (2.13-2.35g/100g) and arginine (1.02-1.04g/100g). These amino acids were also important amino acids in Ab [18], such as glutamic acid and aspartic acid, which were closely related to umami taste [200]. However, the addition of Ab and soybean oil did not change the amino acid profile. Lysine, leucine and arginine were the main essential amino acids in the chicken sausage. Ceron Guevara et al. [201] found that the addition of Ab and *Pleurotus ostreatus* flour as substitutions of salt and fat in frankfurter sausage had no significant effect on the amino acid content of sausage, which was consistent with the results of present study.

Table 4.3 – The amino acids of the chicken sausage with Ab powder and soybean oil

Amino acids	CK	T30	T60	T90
Essential				
Val	0.77±0.01b	0.83±0.02a	0.82±0.01a	0.81±0.01a
Met	0.34±0.01c	0.43±0.02a	0.38±0.02b	0.41±0.02ab

Ile	0.69±0.02c	0.84±0.03a	0.77±0.01b	0.75±0.01b
Leu	1.10±0.05a	1.32±0.21a	1.21±0.10a	1.19±0.08a
Phe	0.61±0.02c	0.72±0.02a	0.66±0.01b	0.65±0.01b
His	0.34±0.01a	0.35±0.02a	0.34±0.01a	0.35±0.02a
Lys	1.18±0.08b	1.37±0.09a	1.26±0.07ab	1.32±0.08ab
The	0.69±0.02b	0.77±0.01a	0.73±0.01a	0.74±0.02a
Arg	1.04±0.08a	1.02±0.05a	1.02±0.04a	1.03±0.03a
Non-essential				
ASP	1.23±0.06b	1.38±0.03a	1.34±0.05ab	1.38±0.04a
Glu	2.13±0.15a	2.29±0.05a	2.33±0.06a	2.35±0.07a
Ser	0.41±0.01c	0.49±0.01a	0.45±0.02b	0.5±0.01a
Gly	0.71±0.01d	0.82±0.01b	0.76±0.02c	0.95±0.01a
Pro	0.55±0.01d	0.64±0.01b	0.59±0.01c	0.69±0.01a
Ala	0.8±0.02c	0.91±0.01a	0.85±0.02b	0.9±0.03ab
Cyst	0.1±0.01b	0.13±0.01a	0.11±0.01ab	0.11±0.01ab
Tyr	0.51±0.02ab	0.53±0.01a	0.49±0.02b	0.52±0.02ab
Total	13.19±0.59b	14.85±0.61a	14.12±0.49ab	14.63±0.48ab
∑EAA	6.76±0.30b	7.65±0.47a	7.19±0.28ab	7.25±0.28ab
∑NEAA	6.44±0.29b	7.19±0.14ab	6.92±0.21b	7.40±0.20a
EAA/NEAA	1.05±0.02b	1.06±0.01a	1.04±0.02ab	0.98±0.02b

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$).

4.2.3 Fatty acid profile of chicken sausage

Table 4.4 shows the fatty acid content of the chicken sausage with Ab powder. The SFA decreased from 36.48 g/100 g in the control group to 31.64 g/100 g in the T30 group, 26.76 g/100 g in the T60 group, and 19.92g/100 g in the T90 group ($P < 0.05$). In fact, except for C20:0 and C22:0, the contents of other fatty acids gradually decreased with the replacement of pork back fat.

Similarly, the content of MUFA decreased ($P < 0.05$), from 48.91 g/100 g in the control group to 43.3, 37.09 and 28.14 g/100 g in T30, T60 and T90, respectively. This decrease is directly related to the low content of C16:1, C17:1, C20:1 and C18:1n9c in the samples containing Ab and soybean oil. In this case, MUFA (except C22:1n9 and C24:1) decreased with the increase of the proportion of Ab and soybean oil, especially the content of C18:1n9c decreased from 45.51g/100g of control samples to 39.81, 34.63, and 26.71 g/100g of T30, T60, and T90, respectively.

Contrary to SFA and MUFA, the total content of PUFA increased significantly with the addition of Ab and soybean oil ($P < 0.05$). In this regard, both C18:2n6 and C18:3n3 were significantly increased, although the contents of C18:3n6, C20:2 and C20:3n3 decreased with the addition of Ab and soybean oil. It is particularly worth mentioning that the content of C18:2n-6 in the samples containing Ab and soybean oil (22.35, 32.81, and 47.03 g/100 g for T30, T60, and T90, respectively) was multiplied compared to the CK (13.00 g/100 g), contributing a large share to the increase in the total PUFA content.

Table 4.4 – The fatty acid of the chicken sausage with Ab powder and soybean oil

Fatty acid	CK	T30	T60	T90
C14:0	1.10±0.23a	0.92±0.14a	0.61±0.12b	0.26±0.21c
C15:0	0.05±0.00a	0.05±0.00a	-	-
C16:0	23.7±2.45a	20.81±0.91a	17.73±1.02b	13.54±1.23c
C17:0	0.27±0.02a	0.24±0.01a	0.19±0.01b	
C18:0	10.91±2.34a	9.43±1.96ab	7.52±1.55ab	5.17±1.02b
C20:0	-	-	0.44±0.01b	0.58±0.02a
C22:0	0.1±0.02d	0.19±0.02c	0.27±0.04b	0.37±0.03a
∑SFA	36.13±2.39a	31.64±2.08b	26.76±2.34c	19.92±2.06d
C16:1	2.2±0.54a	1.93±0.21a	1.29±0.11b	0.59±0.35c
C17:1	0.29±0.01a	0.26±0.02a	0.17±0.04b	0.09±0.00c
C20:1	0.98±0.07a	0.85±0.16a	0.59±0.09b	0.36±0.05c
C18:1n9t	-	-	-	-
C18:1n9c	45.51±3.15a	39.81±2.01b	34.63±2.16c	26.71±3.04d
C22:1n9	0.32±0.03a	0.35±0.02a	0.31±0.04a	0.3±0.01a
C24:1	0.1±0a	0.1±0.00a	0.09±0.01a	0.09±0.01a
∑MUFA	49.41±3.12a	43.3±2.45b	37.09±2.46c	28.14±2.37d
C18:2n6	13.00±1.04d	22.35±2.36c	32.81±2.41b	47.03±2.17a
C18:3n6	0.27±0.08a	0.38±0.04a	-	-
C18:3n3	0.6±0.04d	1.82±0.11c	3.05±0.63b	4.78±1.04a
C20:2	0.53±0.08a	0.45±0.07a	0.29±0.02b	0.13±0.01c
C20:3n3	0.06±0.01a	0.06±0.00a	-	-
∑PUFA	14.47±2.31d	25.06±3.05c	36.15±3.14b	51.94±3.48a
PUFA/SFA	0.4±0.02d	0.79±0.07c	1.35±0.09b	2.61±1.01a
PUFA/MUFA	0.29±0.02d	0.58±0.11c	0.97±0.14b	1.85±0.19a
MUFA/SFA	1.37±0.03a	1.37±0.09a	1.39±0.08a	1.41±0.14a
(MUFA+PUFA)/SFA	1.77±0.09d	2.16±0.11c	2.74±0.24b	4.02±0.85a

Note: All values are the mean ± standard error; Saturated fatty acids (SFA); Polyunsaturated fatty acids (PUFA); Monounsaturated fatty acids (MUFA). Different lowercase letters mean significant differences in the same line ($P < 0.05$).

4.2.4 CY

As can be seen from Figure 4.5 that the cooking yield of chicken sausage with Ab mushrooms and soybean oil as a substitute for fat has no significant difference ($P < 0.05$) compared with the control and was close to 100%. One reason might be that soybean oil had a good emulsifying ability. Many studies have confirmed that replacing fat with vegetable oil could improve cooking yield. In addition, Ab powder is rich in dietary fibre, which can absorb water and oil and reduce the cooking loss of meat products. Dietary fibre has been proved a good fat substitute. In addition, Ab mushrooms contained up to 22.14% protein (Table 4-1), which can also improve the emulsifying effect of meat. Therefore, the cooking yield of sausage with Ab mushrooms and soybean oil as fat substitute did not decrease.

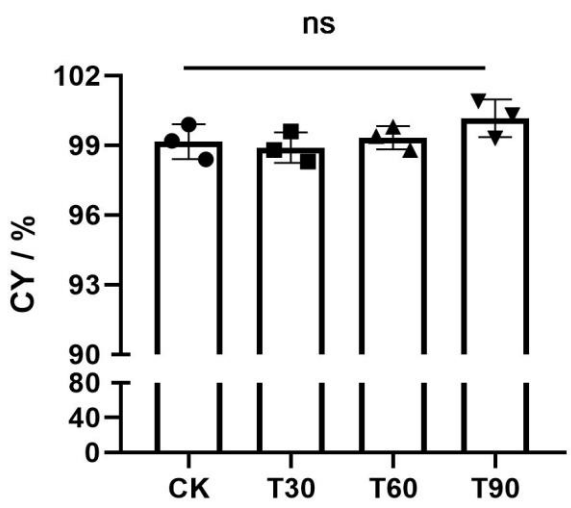

Figure 4.5 – Cooking yield of the chicken sausage.

Note: CK, T30, T60 and T90 represent 0 %, 30 %, 60 % and 90 % fat replacement in sausage, respectively. ns indicates no significant differences between the treatments (P > 0.05).

4.2.5 Microstructure

The microstructures of chicken sausage with Ab and soybean oil as fat substitute are presented in Figure 4.6. It can be seen from Figure 4.6 that the microstructure of the chicken sausage gel had changed significantly with the addition of Ab and soybean oil. The CK presented a granular rough appearance. However, the microstructures of T30, T60, and T90 showed fewer cavities and were more uniform and denser than the CK. Moreover, the greater the amount of Ab and soybean oil incorporated, the fewer cavities and denser appearance appeared. There may be many reasons for this result. First, the dietary fibre in Ab had a large molecular structure, which formed more bridges between adjacent macromolecules and promoted the formation of a protein gel network. Secondly, the insoluble substances in Ab powder filled the three-dimensional network structure formed by myofibrillar protein. Thirdly, Ab and soybean oil were pre-mixed, and the soybean oil was absorbed by the small Ab particles, making it evenly dispersed in the Ab particles, which was equivalent to the pre-emulsification of soybean oil. In this way, the soybean oil would be better dispersed with the Ab powder during the process of meat batters production, reducing the formation of large oil droplets, promoting oil droplets to be wrapped with more myofibrillar protein, facilitating the formation of a more consistent gel. Thereby the texture characteristics of the sausage were correspondingly improved.

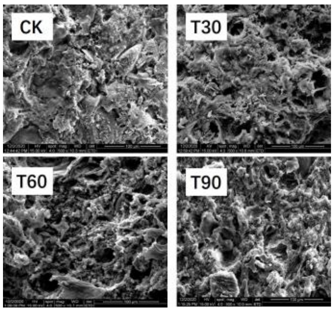

Figure 4.6 – Microstructure of the chicken sausage with Ab powder and soybean oil

4.2.6 Color

The L*, a*, and b* value of the chicken sausage are shown in Table 4.5, Table 4.6, and Table 4.7. L* value represents brightness. During the storage period, the L* value of all samples showed a downward trend in general, and in the same storage period, the L* value decreased significantly with the increase of the fat replacement amount ($P < 0.05$), which indicated that Ab and soybean oil significantly affected the brightness of chicken sausage. The L* value of the control group began to decrease after 20 days of storage, while the L* value of the T30, T60 and T90 groups began to decrease after the 15th day of storage, which may be due to the color change caused by the instability of pigment substances in Ab mushroom.

a* value represents the degree of redness, and the larger the a* value, the more vivid red the product is. It can be seen from Table 4.6 that the incorporation of Ab powder and soybean oil significantly increased a* value of sausage ($P < 0.05$). The redness values of chicken sausage increased significantly with the increase of the content of Ab powder and soybean oil ($P < 0.05$), which may be due to the higher a* value of Ab powder than the CK (Table 5.1). During the storage period, a* values of the control group gradually decreased. On the contrary, a* values of the T30, T60 and T90 groups increased gradually, the possible reason was that the pigment from the Ab mushroom gradually decomposed, resulting in a product with a high a* value. The mechanism of Ab color change needs further study.

b* value represents the yellowness. The b* value of the CK was relatively stable during the storage period and increased after 25 days of storage. The b* values of T30, T60 and T90 were higher than the control in the first five days of storage and decreased during the storage period. On the 10th day of storage, b* value was lower than that of the control. It is worth noting that the contents of Ab and soybean oil in the sausage were higher, the lower the b* value, the faster the b* value decreases. The reason may be that the pigment in Ab was unstable and decomposed during storage, producing substances with lower yellowness.

In conclusion, the color of the sausage changed during storage. Its brightness decreased. The redness of the CK decreased and the yellowness value increased. The addition of Ab and soybean oil increased the redness and decreased the yellowness of sausage. The reason may be that the colored substances produced by browning of Ab mushroom during the drying process were unstable, and decomposed into substances with high redness and low yellowness.

Table 4.5 –L* value of the chicken sausage with Ab powder and soybean oil

Day/d	L*			
	CK	T30	T60	T90
1	86.78±0.06a,A	75.52±0.23b,A	68.42±0.39c,A	64.20±0.15d,A
5	86.60±0.16a,A	75.1±0.37b,A	68.98±0.18c,A	64.54±0.37d,A
10	86.47±0.1a,A	74.79±0.10b,A	68.64±0.08c,A	64.06±0.16d,A
15	86.45±0.36a,A	74.07±0.03b,B	67.47±0.10c,B	62.49±0.18d,B
20	85.71±0.25a,B	73.10±0.44b,C	67.05±0.08c,C	62.32±0.22d,BC
25	85.32±0.26a,B	72.09±0.32b,D	65.72±0.02c,D	62.20±0.46d,C
30	84.06±0.05a,C	72.33±0.15b,D	65.50±0.45c,D	60.38±0.05d,D
35	81.12±0.19a,D	69.83±0.30b,E	63.51±0.21c,E	58.39±0.26d,E

Note: Different lowercase letters mean significant differences in the same line ($P < 0.05$). Different capital letters mean significant differences in the same column ($P < 0.05$).

Table 4.6 –a* value of the chicken sausage with Ab powder and soybean oil

Day/d	a*			
	CK	T30	T60	T90
1	0.43±0.01d,A	1.24±0.02c,F	2.4±0.02b,G	2.65±0.03a,E
5	0.37±0.06d,AB	1.23±0.04c,E	2.38±0.02b,G	2.87±0.07a,D
10	0.33±0.01d,B	1.25±0.01c,E	2.48±0.08b,F	2.97±0.03a,D
15	0.27±0.02d,B	1.37±0.03c,D	2.61±0.08b,E	3.44±0.08a,C
20	0.14±0.02d,C	1.43±0.01c,C	2.74±0.01b,D	3.50±0.01a,C
25	0.11±0.01d,C	1.47±0.03c,C	2.86±0.03b,C	3.58±0.04a,B
30	0.12±0.02d,C	1.92±0.03c,B	3.10±0.03b,B	3.65±0.03a,B
35	0.08±0.01d,D	2.23±0.02c,A	3.41±0.06b,A	4.23±0.02a,A

Note: Different lowercase letters mean significant differences in the same line ($P < 0.05$). Different capital letters mean significant differences in the same column ($P < 0.05$).

Table 4.7 –b* value of the chicken sausage with Ab powder and soybean oil

Day/d	b*			
	CK	T30	T60	T90
1	14.25±0.15d,B	17.47±0.09a,A	16.82±0.08b,A	16.29±0.02c,A
5	14.62±0.45b,B	15.78±0.14a,B	15.52±0.16a,B	15.54±0.09a,B
10	14.93±0.13a,B	14.69±0.02b,C	14.44±0.15c,C	14.25±0.02d,C
15	14.94±0.09a,B	14.63±0.04b,C	13.75±0.15b,D	14.07±0.09c,D
20	14.53±0.45a,B	14.72±0.23a,C	13.77±0.02b,D	13.53±0.10c,E
25	15.36±0.13a,A	14.75±0.04b,C	13.73±0.02b,D	13.57±0.05c,E
30	15.35±0.08a,A	13.94±0.04b,D	13.69±0.07c,D	13.57±0.08c,E
35	15.78±0.38a,A	13.86±0.10b,D	13.51±0.15c,D	13.61±0.04c,E

Note: Different lowercase letters mean significant differences in the same line ($P < 0.05$). Different capital letters mean significant differences in the same column ($P < 0.05$).

4.2.7 TPA

Table 4.8 showed the effect of adding Ab powder and soybean oil on the firmness, springiness, cohesiveness and chewiness of chicken sausages. It can be seen from Table 4.8 that the hardness of the sausage increased significantly when adding Ab powder and soybean oil to replace the back fat ($P < 0.05$), and the more the amount of addition, the higher the hardness. During the storage period, although the hardness value of the CK fluctuated, it still showed a general increase and reached the maximum at 35 days of storage. The hardness of the T30 and T60 group gradually increased and reached a maximum at 35th and 10th day of storage respectively. The values of the T90 group reached the maximum on the 20th day of storage, and then remained stable.

It can be seen from Table 4.9 that the addition of Ab powder and soybean oil replaced fat had no significant effect on the springiness of chicken sausage. During storage, the springiness of the control group did not change significantly, the springiness of the T30 group decreased slightly after 35 days of storage, and the springiness of the T60 and T90 groups fluctuated during the storage period and decreased after the 35th day of storage. When stored for 35 days, there was no significant difference in springiness among the four groups of samples.

It can be seen from Table 4.10 that the addition of Ab powder and soybean oil had no significant effect on the cohesiveness of chicken sausage except for T90 group. During storage, the cohesiveness of the control group first decreased and then increased, the cohesiveness of the T30 and T60 groups remained unchanged, while the cohesiveness of the T90 group gradually increased. At the beginning of storage, there was little difference in cohesiveness among the groups. Nevertheless, the cohesiveness of the control group was significantly higher than that of T30, T60 and T90 after 25 days of storage. The changes in cohesiveness showed that an appropriate amount of fat replacement was helpful to maintain the cohesiveness of chicken sausage.

As can be seen from Table 4.11, the chewiness of the T30 group was not significantly different from that of the CK. Although there was no significant difference in the chewiness of the T60 and T90 groups, they were both significantly higher than the CK ($P < 0.05$). The chewiness of the CK did not change significantly during the first 20 days of storage, while the chewiness of the T60 and T90 groups fluctuated to a certain extent during the storage period, reaching the maximum on the 10th and 20th day of storage respectively. Overall, the addition of Ab powder and soybean oil significantly increased the chewiness of the chicken sausage, and the chewiness of sausage increased with the increase of the amount of Ab powder.

Table 4.8 – The hardness of the chicken sausage with Ab powder and soybean oil

Day/d	Hardness/g			
	CK	T30	T60	T90
1	4332.38±190.69b,CD	4414.24±296.9b,C	5810.04±593.49a,E	6301.8±329.22a,E
5	4439.57±356.25c,BC	4612.44±262.46c,C	5978.36±54.74b,DE	6845.19±185.91a,D
10	4302.39±19.6c,CD	5397.42±119.71b,B	7360.61±94.12a,A	7240.68±232.04a,BCD
15	4289.58±120.75b,CD	4479.36±271.15b,C	6753.28±90.01a,BC	6989.81±212.24a,CD

20	4694.87±12.16c,B	5074.61±412.61c,B	6175.26±77.56b,DE	7879.9±469.98a,A
25	4042.96±59.05c,DE	5070.86±220.84b,B	6414.66±27.8a,BCD	6346.66±175.42a,E
30	3848.07±203d,E	5153.51±108.92c,B	6310.01±252.04b,CD	7611.23±43.41a,AB
35	5167.46±192.17d,A	6036.76±202.86c,A	6823.61±340.14b,B	7458.83±207.57a,ABC

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$). Different capital letters mean significant differences in the same column ($p < 0.05$).

Table 4.9 – The springiness of the chicken sausage with Ab powder and soybean oil

Day/d	Springiness			
	CK	T30	T60	T90
1	0.931±0.003a,A	0.931±0.003a,A	0.942±0.012a,AB	0.932±0.006a,AB
5	0.927±0.005ab,A	0.928±0.005ab,A	0.948±0.013a,A	0.923±0.015b,ABC
10	0.926±0.021a,A	0.933±0.004a,A	0.922±0.001a,CD	0.937±0.004a,A
15	0.925±0.007ab,A	0.933±0.005a,A	0.915±0.005c,D	0.918±0.003bc,C
20	0.921±0.010b,A	0.934±0.003a,A	0.918±0.002b,D	0.921±0.002b,BC
25	0.922±0.004b,A	0.931±0.005a,A	0.934±0.004a,BC	0.914±0.003c,C
30	0.93±0.002b,A	0.927±0.003bc,A	0.935±0.003a,ABC	0.924±0.003c,ABC
35	0.921±0.002a,A	0.915±0.006a,B	0.914±0.006a,D	0.915±0.012a,C

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$). Different capital letters mean significant differences in the same column ($p < 0.05$).

Table 4.10 – The cohesiveness of the chicken sausage with Ab powder and soybean oil

Day/d	Cohesiveness			
	CK	T30	T60	T90
1	0.761±0.004a,AB	0.720±0.024ab,A	0.739±0.018a,A	0.697±0.016b,CD
5	0.733±0.01a,C	0.729±0.012a,AB	0.733±0.013a,A	0.698±0.003b,CD
10	0.734±0.003a,C	0.704±0.003c,C	0.717±0.006b,A	0.704±0.005c,BC
15	0.735±0.011a,C	0.733±0.005a,AB	0.72±0.001b,A	0.708±0.003c,BC
20	0.712±0.006c,D	0.748±0.005a,A	0.726±0.005b,A	0.692±0.005d,D
25	0.772±0.009a,A	0.74±0.003b,A	0.726±0.015bc,A	0.722±0.003c,A
30	0.76±0.008a,AB	0.739±0.009ab,A	0.729±0.019b,A	0.698±0.002c,CD
35	0.752±0.002a,B	0.715±0.004b,BC	0.724±0.012b,A	0.713±0.002b,AB

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$). Different capital letters mean significant differences in the same column ($p < 0.05$).

Table 4.11 – The chewiness of the chicken sausage with Ab powder and soybean oil

Day/d	Chewiness/g			
	CK	T30	T60	T90
1	3048.55±100.41b,B	3030.04±80.84b,C	3896.93±152.17a,E	4074.98±72.42a,D
5	3018.52±242.78c,B	3037.79±103.09c,C	3978.34±109.29b,DE	4374.35±77.56a,C
10	2904.51±69.93c,BC	3680.23±175.05b,B	4843.97±67.42a,A	4680.65±71.5a,B
15	2887.58±45.69c,BC	3019.75±233.29c,C	4230.46±132.55b,BC	4767.4±103.84a,B
20	3044.03±60.86d,B	3497.78±285.16c,B	4153.87±59.68b,CD	5041.49±298.61a,A
25	2767.35±46.15c,C	3622.5±112.44b,B	4258.21±12.53a,BC	4248.72±93.83a,CD
30	2733.9±178.64d,C	3476.99±6.79c,B	3890.39±187.10b,E	4865.76±37.3a,AB

35	3535.13±128.15d,A	3971.81±170.16c,A	4393.43±142.27b,B	4729.74±138.71a,B

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$). Different capital letters mean significant differences in the same column ($p < 0.05$).

4.2.8 pH

As can be seen from Table 4.12 that the pH values of the T60 and T3 groups were significantly higher than those of the control group as expected ($P < 0.05$). The reason for this should be that the pH values of Ab and soybean oil were higher than that of the fat. During storage, the pH values of both CK and T30 groups began to decrease on the 25th day of storage, and the pH values of the T60 group did not change significantly during the whole storage period. Notably, the pH value of the T90 group decreased sharply to 6.36 on the 35th day of storage, but it was still significantly higher than that of the CK ($P < 0.05$). The possible reason for this may be that there was the highest content of soybean oil and PUFA (Table 4.4) in the T90, which were very unstable and more prone to oxidative rancidity[202]. FL á via Santi stefanello et al.[203] also found similar changes in pH value about fresh pork sausage containing sun mushroom powder. The pH change of meat products during storage was related to microbial growth. The decrease in pH may be due to the accumulation of endogenous or acid-producing microorganisms, such as lactic acid bacteria. Longer storage time increased the number of lactic acid-producing microorganisms, causing the pH to drop more.

In the early stage of sausage storage, since most of the microorganisms were killed during the heat treatment, the growth ability of some of the surviving bacteria had not been recovered. In addition to the inhibition of low temperature and vacuum conditions, the growth of bacteria was slow, which decomposed sugar substances and generated lactic acid, acetic acid, etc. Weak organic acids were less capable, so pH did not change significantly. At the later stage of storage, due to temperature fluctuations, some of the surviving bacteria, especially the residual lactic acid bacteria, recovered their growth ability and used the rich nutrients in sausages to accumulate lactic acid. Thereby, the pH value of sausages decreased. When stored for 35 days, the pH values of T30, T60, and T90 were higher than that of the CK, indicating that Ab inhibited the growth of bacteria to a certain extent.

Table 4.12 – The pH values of the chicken sausage with Ab powder and soybean oil

Day/d	pH			
	CK	T30	T60	T90
1	6.43±0.01c,A	6.47±0.03bc,A	6.48±0.03b,A	6.53±0.01a,A
5	6.43±0.03c,A	6.49±0.01b,A	6.48±0.01b,A	6.53±0.01a,A
10	6.47±0.05b,A	6.50±0.01b,A	6.49±0.02b,A	6.53±0.01a,A
15	6.47±0.04b,A	6.52±0.00a,A	6.53±0.02a,A	6.54±0.02a,A
20	6.48±0.04b,A	6.51±0.02b,A	6.52±0.03ab,A	6.56±0.02a,A
25	6.36±0.03c,B	6.43±0.01b,B	6.45±0.02b,A	6.54±0.01a,A
30	6.32±0.02c,B	6.44±0.00b,B	6.49±0.01a,A	6.50±0.01a,A
35	6.23±0.03c,C	6.35±0.01b,C	6.49±0.02a,A	6.36±0.02b,B

Note: Different lowercase letters mean significant differences in the same line ($p < 0.05$). Different capital letters mean significant differences in the same column ($p < 0.05$).

4.2.9 TBARS value

It can be seen from Table 4.13 that the TBARS values of the CK and T30 increased, reached the maximum on the 25th day of storage at 4 °C, and then decreased. TBARS in the T60 and T90 groups first decreased and then increased from 0 to 15 days, reached a maximum at 25 days of storage, and then decreased.

It is worth noting that the TBARS values of T30, T60, and T90 were higher than those of the CK in the first 5 days of storage. The reason for this might be that the browning reaction and protein degradation products of Ab mushroom during drying, as well as the pigments contained in soybean oil participated in the formation of thiobarbituric acid (TBA) color complex, thereby overestimated the malondialdehyde (MDA) value.

It is reported that the antioxidant substances in Ab mushroom are phenolic acids, especially the hydroxy derivatives of benzoic acid [204]. The present studies showed that Ab and soybean oil as compound fat substitutes did reduce the lipid oxidation of chicken sausage. Alnoumani et al. [205] found that dry Ab powder inhibited lipid oxidation of cooked beef, which was consistent with the results of present study. Nevertheless, some studies have found that the addition of Ab did not significantly inhibit the lipid oxidation of meat products [201]. The reason for this might be that there were great differences in the composition and functional components of Ab mushroom under different growth, harvest and processing conditions [206].

During storage, the increase or decrease of TBARS value had certain volatility, indicating that fat oxidation was not the main factor of chicken sausage spoilage. The possible reason was that fat particles were very small and wrapped by soluble protein, so they were not so easy to be oxidized or decomposed by microorganisms.

Table 4.13 – The TBARS value of the chicken sausage with Ab powder and soybean oil

Day/d	TBARS/ (mg/100g)			
	CK	T30	T60	T90
1	0.101±0.001d,D	0.148±0.001c,E	0.202±0.001b,C	0.275±0.001a,A
5	0.147±0.004d,C	0.156±0.001c,D	0.185±0.003b,D	0.261±0.006a,B
10	0.217±0.013a,B	0.169±0.006b,C	0.174±0.003b,E	0.227±0.003a,C
15	0.291±0.003a,B	0.181±0.007b,B,C	0.159±0.004c,F	0.150±0.005c,E
20	0.295±0.001a,B	0.189±0.001bc,B	0.235±0.006b,B	0.181±0.001c,D
25	0.328±0.006a,A	0.224±0.003c,A	0.247±0.005b,A	0.223±0.002c,C
30	0.298±0.006a,B	0.210±0.013b,A	0.159±0.006c,F	0.102±0.008d,G
35	0.232±0.002a,C	0.161±0.002b,C	0.147±0.003c,G	0.126±0.001d,F

Note: Different lowercase letters mean significant differences in the same line ($P < 0.05$). Different capital letters mean significant differences in the same column ($P < 0.05$).

4.2.10 Total viable counts (TVC)

TVC values in chicken sausage during the 35 days storage at 4°C shown in Table 4.14. As shown in Table 4.14, the addition of Ab and soybean oil had a significant ($P < 0.05$) effect on microbial growth. The total initial colonies of the CK, T30, T60, and T90 were 212, 37, 27, and 37 CFU/g, respectively. The number of microorganisms in the sausages of the CK increased slowly in the early stage of storage and gradually increased in the later stage. The lower TVC value in the early stage of refrigeration may be related to low temperature, and the stress condition of low temperature may lead to a lower bacterial metabolic rate. On the 15th day of storage at 4°C, the TVC value in CK continued to rise, reaching about 4.50×10^4 CFU/g on the 30th day, which did not exceed the limit of the Chinese national standard (10^5 CFU/g). However, it surged to 1.65×10^8 CFU/g on the 35th day of storage, which was a ten thousand-fold surge compared with the value on the 30th day, indicating that the bacteria entered the logarithmic growth phase at this time.

The TVC value in the three groups containing Ab and soybean oil was significantly lower than that of the control group at 0 days of storage, and there was no significant difference among these three groups ($P < 0.05$), which may be related to the inhibitory effect of Ab on spoilage bacteria. This antibacterial activity was related to the main active ingredients, such as catechin, caffeic acid and rutin. Catechins, a phenolic substance, has been proved to have antibacterial activity [63]. Caffeic acid and rutin have been shown to have antibacterial activity as well[123].

However, the TVC value remained at 1.43×104-1.78×10^5 CFU/g from the 5th day of storage, which was much higher than that of the CK, and remained stable during the 25-day cold storage period. Cerón-Guevara et al. studied the TVC value of refrigerated frankfurter sausage [201] and liver pâté [207] with Ab mushroom and *Pleurotus ostreatus* powder and found that the TVC value of the treatment group was much higher than that of the CK, which was consistent with the results of the present study. For low-temperature meat products, heat treatment can only kill the vegetative body of microorganisms, and the more heat-resistant spores, fungal spores and some heat-resistant microorganisms can not be completely eliminated. Therefore, these high levels of microorganisms can be attributed to the spore-forming bacteria naturally present in Ab mushroom, which survived to thermal treatment. During storage, it was observed that the sausage containing Ab powder and soybean oil had no peculiar smell and bagging phenomenon during 35 days of storage. Combined with the changes of sausage texture, pH and TBARS value during storage, this high TVC value was not considered a risk [201]. During the 30 to 35 days of storage, the TVC value of T30, T60, and T90 increased by a hundred times, which may be related to the increase in the number of psychrophilic bacteria in the sausage, but the growth rate was still lower than that of the CK, which increased by ten thousand times. The TVB value of CK at 30 days of storage exceeded the Chinese national standard limit value. Therefore, it can be concluded that the storage period of sausages containing Ab mushroom and soybean oil is at least 25 days.

Table 4.14 – The TVC value of the chicken sausage with Ab powder and soybean oil

Day/d	Total viable counts / (CFU/g)			
	CK	T30	T60	T90
0	2.12×10^2±1.32a,F	3.7×10±0.6b,F	2.7×10±0.5b,D	3.7×10±0.5b,D
5	2.9×10^2±1.02c,F	3.9×10^4±1.12a,E	1.43×10^4±1.09b,C	1.59×10^4±1.21ab,C
10	5×10^2±1.21b,F	5.25×10^4±1.33a,E	3.00×10^4±1.23a,C	2.75×10^4±1.32a,C
15	2.25×10^3±0.98c,E	7.20×10^4±1.45a,DE	4.15×10^4±1.36b,C	5.10×10^4±1.26b,C
20	8×10^3±1.24c,D	9.15×10^4±1.26a,D	7.90×10^4±1.42a,B	1.20×10^5±1.00b,C

25	$2.1\times10^4\pm1.01$b,C	$1.78\times10^5\pm1.01$a,C	$8.25\times10^4\pm1.21$a,B	$1.09\times10^5\pm0.93$a,C
30	$4.5\times10^4\pm1.25$c,B	$1.89\times10^6\pm1.11$b,B	$1.98\times10^6\pm1.15$b,B	$2.08\times10^7\pm1.05$a,B
35	$1.65\times10^8\pm0.95$b,A	$1.90\times10^9\pm1.42$a,A	$3.00\times10^9\pm1.06$a,A	$1.28\times10^9\pm0.97$a,A

Note: Different lowercase letters mean significant differences in the same line ($P < 0.05$). Different capital letters mean significant differences in the same column ($P < 0.05$).

4.3 Technological schemes of the sausages production

By optimizing the formula of sausage, the optimal formula obtained of the low-fat chicken sausage is as follows:

60 g of chicken breast meat, 8 g of pork back fat, 4 g of Ab powder, 8 g of soybean oil, 20 g of ice water, 1.4 g of refined salt, and 0.3 g of tripolyphosphate, 0.1 g of chicken essence, 0.65 g of sugar, 0.15 g of white pepper, 5 g of potato starch.

The sausage production process is shown in Figure 4.7.

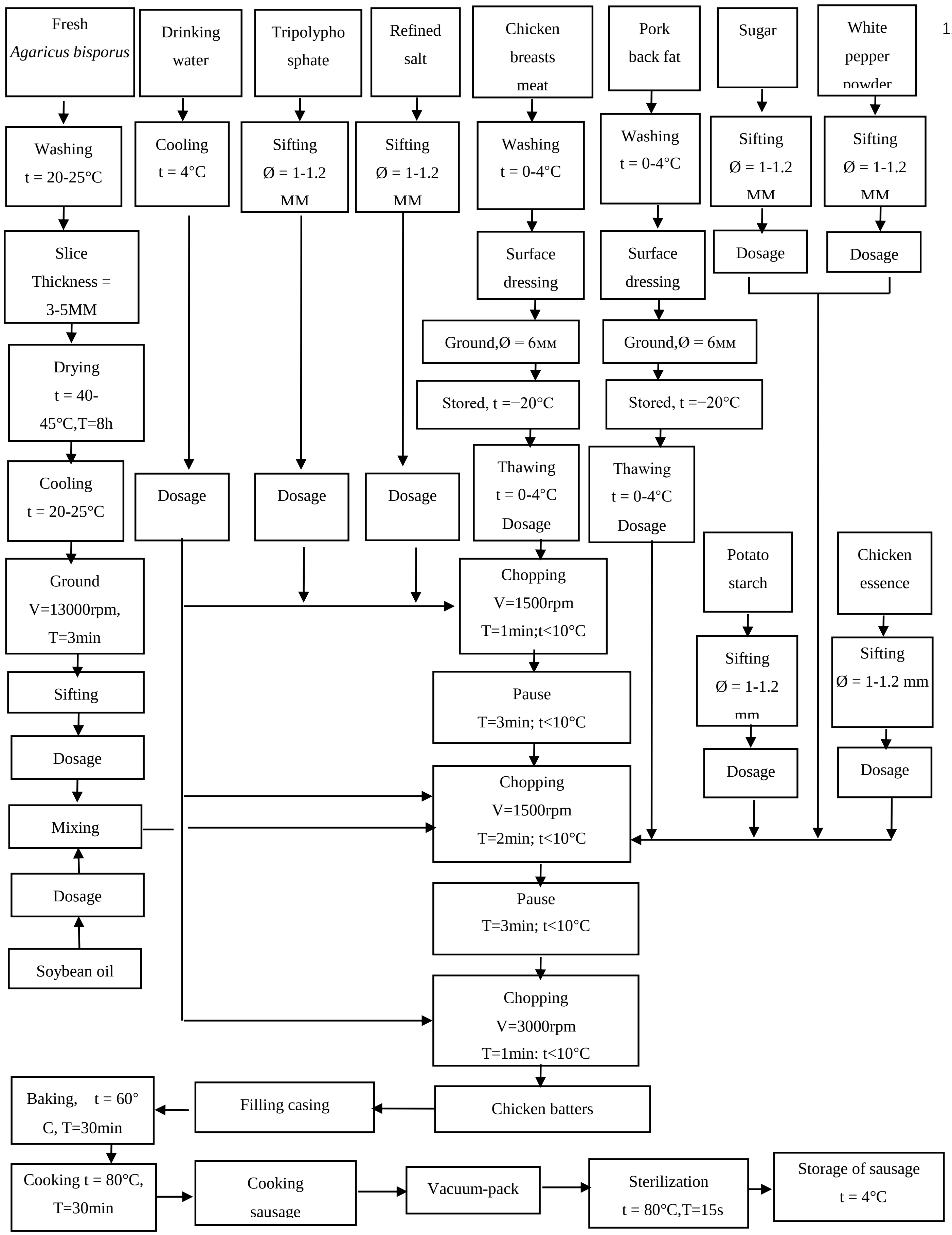

Fig 4.7 – Process flow chart of chicken sausage production

CONCLUSION OF SECTION 4

1. Low-fat chicken sausage with Ab and soybean oil was created. Sensory evaluation was used to optimize the amount of potato starch, chicken essence, refined salt added, and the premix of Ab and soybean oil. The nutritional, sensory, manufacturing, and storage characteristics of the sausage with the premix of Ab and soybean oil were evaluated. The technological schemes of the low-fat chicken sausages with Ab are determined.

2. The addition of potato starch had a greater effect on the texture, taste and general acceptability of the chicken sausage, and the addition of 5% potato starch showed the best sensory properties.

3. Chicken essence had a greater effect on the texture and taste of the chicken sausage, and 0.1% chicken essence showed the best sensory properties.

4. The amount of refined salt added had a greater impact on the texture, flavor and general acceptability of the chicken sausage, and the addition of 1.4% of the refined salt showed the best sensory properties.

5. Ab mushroom and soybean oil improved the taste, texture, and overall acceptability of the sausage. The sausage with a 60% fat replacement percentage owned the finest sensory characteristics.

6. With the increase of Ab and soybean oil content, the amount of fat and energy in the sausage greatly decreased, the amount of ash and carbohydrates significantly rose. The cooking yields of the sausages were consistent and were all very close to 100%. The protein levels of the sausages decreased dramatically when 90% of the fat was replaced. The content of essential amino acids of the sausage with the 30% fat replacement percentage dramatically increased. The amino acid profile in this scenario was unaffected greatly by the replacement quantities of 60% and 90% fat. The amount of SFA and MUFA in chicken sausages dramatically dropped, although the amount of PUFA, PUFA/MUFA, and (PUFA + PUFA)/SFA greatly rose. Comprehensive investigation revealed that the optimum nutritional characteristics of the sausage came from a fat replacement ratio of 60%.

7. Low-fat chicken sausages showed a denser, more homogeneous, and fewer

cavities microstructure than high-fat chicken sausages thanks to the addition of Ab and soybean oil as fat substitutes.

8. Color and color preservation properties of the sausage were altered by Ab and soybean oil. The L* value of the low-fat chicken sausage dramatically dropped, while a* and b* values rose significantly. The a* value steadily increased while the L* and b* values gradually decreased during storage. While the L* and a* values of the high-fat sausage gradually declined, b* value gradually rose during the storage.

9. The hardness and chewiness of chicken sausages significantly improved when 60 % and 90 % of the fat were substituted by Ab and soybean oil. Overall hardness of the low-fat chicken sausage increased, springiness and chewiness fluctuated during storage. Low-fat chicken sausage with a 60% fat replacement percentage maintained its cohesiveness throughout the storage period.

10. The pH value of the low-fat chicken sausage containing Ab and soybean oil increased and was relatively stable during storage. The greater the amount of Ab added, the greater the pH value. The pH of the sausage with 60% fat replacement percentage had no change throughout the storage period.

11. The TBARS values of the low-fat chicken sausages increased noticeably during storage, and their range of TBARS values was narrower than that of high-fat chicken sausages. The TBARS values of low-fat chicken sausages were significantly lower than high-fat chicken sausages on day 15 of storage. The lower the TBARS value, the higher the level of Ab mushroom and soybean oil. Ab mushroom partially reduced the oxidation of fat in chicken sausage.

12. The total number of colonies in high-fat chicken sausage exceeded the Chinese national standard after 35 days of storage. Only on the first day was the total number of colonies for the low-fat chicken sausages lower than that of the high-fat chicken sausages. Thereafter, the number of colonies remained high and slowly climbed throughout the storage period. Low-fat chicken sausages containing Ab and soybean oil had a minimum shelf life of 25 days.

SECTION 5 ECONOMIC BENEFIT ANALYSIS

The purpose of the work was to produce a low-fat chicken sausage containing Ab at a food processing enterprise. Each shift has a capacity of 200kg to meet the needs of the population by creating an economically efficient production of them for a stable income.

5.1 Production program

The calculation of the production program is presented in Table 5.1.

Table 5.1 – Production volume in value terms

Product type	Production volume per shift, kg	Price of sold products, Dollar.
Dough	200	**912.8**
Together		**912.8**

5.2 The cost of raw materials, basic materials, and auxiliary materials

5.2.1 The cost of raw materials, basic materials

In order to ensure the production of products that meet modern requirements, the workshop does not need a radical reconstruction both in terms of replacing outdated equipment and in terms of introducing modern technologies.

Based on the calculations of the technology project and the data of technological practice, we calculate raw materials and basic materials for the production of a low-fat chicken sausage.

Table 5.2 - Calculation of the cost of raw materials and basic materials

Type of raw material	The need for raw materials per shift, kg	Purchase price for 1 kg, Dollar.	Total cost of raw materials, Dollar.
chicken breast	111.5	2.43	270.56
pork back fat	14.9	3.57	53.17
Ab powder	7.4	13.99	103.51
soybean oil	14.9	1.38	20.50
ice water	37.2	0.0001	0.01
refined salt	2.6	0.89	2.32
Tripolyphosphate	0.6	1.43	0.86
chicken essence	0.2	5.55	1.11
sugar	1.2	1.22	1.46

white pepper	0.3	14.79	4.44
potato starch	9.3	3.40	31.59
Totaly	200		489.52

5.2.2 The cost of auxiliary materials

Next, we determine the amount and cost of auxiliary materials for the production of low-fat chicken sausage. At the same time, only the cost of auxiliary materials for technological purposes is taken into account, which is calculated directly, based on the costs of the entire production and the cost of auxiliary materials.

(1) Collagen casing: 18mm diameter collagen casing is used for sausage filling. During filling, the consumption of collagen casings with a diameter of 18 mm is 6.83m per kilogram of meat filling, and the casing loss is 0.1%.

The consumption of collagen casings per shift is 200×6.83×(1+0.1%)=1502.6 meters.

(2)Packaging carton: five-layer corrugated cardboard is used. Each sausage weighs 0.021kg, and 50 pieces are packed in each carton, with a loss of 0.1%.

The number of cartons required for each batch is 200 ÷ 0.021 ÷ 50 × (1+0.1%)=191.

(3)Tape: carton size is 280 × 195 × 105mm, extending 1cm at both ends, loss is calculated as 0.1%.

The length of tape required for each carton is (280+10 × 2) × 2=600mm.

Daily consumption of adhesive tape length is 191×600×(1+0.1%)÷1000=115m.

(4) Tensile film: loss is calculated as 0.2%. 0.045kg of base film is consumed per kilogram of sausage, 0.03kg of covering film is consumed per kilogram of sausage.

The number of base film required per day is 200×0.045×(1+0.2%)=9.02kg.

The number of covering film per day is 200 × 0.03 × (1+0.2%)=6.02kg.

The calculations are presented in Table 5.3.

Table 5.3 - Calculation of the cost of auxiliary raw materials and materials

Type of raw material	Need for materials, number	Purchase price, Dollar	Total cost, Dollar
Collagen casing	1502.6 meters	0.06/meter	92.22
Packaging carton	191PCS	0.31/PCS	59.98
Tape	115meters	0.0049/meter	0.57
Tensile film(base film)	9.02kg	4.43/kg	39.91

Tensile film(covering film)	6.02kg	5.00/kg	30.07
Together			130.54

5.3 The salary of employees

We will calculate the salary of employees.

Table 5.4 - Calculation of the salary fund

Type of employee	Number of employees	Basic salary, Dollar	Additional salary, Dollar.	Charges for wages (37.5%), Dollar.	Basic salary fund, Dollar
10	10	32.52	3.25	13.41	49.19

5.4 The cost of fuel and energy

The calculation of energy consumption is carried out based on the norms of consumption of energy resources per one ton of product and their cost, based on the data of the energy and electrical engineering calculation of the project, which are given in the feasibility study.

Table 5.5 - Calculation of the cost of fuel and energy for the production of products

Types of fuel and energy	Rate per 1000 kg of products	The number of production per shift	Price per unit, Dollar.	Total costs Dollar
Electricity	575.65 kW	0.2	0.05	6.02
Water	7.74 m^3	0.2	0.35	0.55
In total	h	h	h	6.57

5.5 The costs of equipment maintenance and operation

The costs of maintenance and operation of machines and equipment are determined depending on the complexity of the innovative solution:

we accept in the amount of 20% of the basic salary in the absence of capital investments=6.50 Dollar.

5.6 Total expenditures

General production costs are accepted in the amount of 50% of the basic salary=16.26 Dollar.

5.7 Administrative costs

Administrative costs are 1.5% of the production cost of products=10.39Dollar.

5.8 Sales costs

Sales costs are 10% of the production cost of products =69.26Dollar.

5.9 Other operating costs

Other operating costs are 5% of the production cost of products = 34.63Dollar

After the calculations, a summary table of production costs is drawn up.

Table 5.6 – The cost of production

No	Articles of expenditure	Sum, Dollar
1	Raw materials and materials	**489.52**
2	Auxiliary materials	130.54
3	Fuel and energy for technological purposes	6.57
4	Salary with deductions	49.19
5	Equipment maintenance and operation costs	6.50
6	Total expenditures	16.26
7	Production cost	**692.62**
8	Administrative expenses	10.39
9	Selling expenses	69.26
10	Other expenses	34.63
11	Full cost	**806.90**

5.10 Technical and economic performance indicators of the enterprise

Summarizing indicators of the company's activity are given in the table 5.7.

Table 5.7 - Technical and economic performance indicators of the enterprise

Indicators	Unit	Value
Volume of produced products in current prices	Dollar	913.51
Full costs of production and sale of products	Dollar	806.90
Expenses for 1 hryvnia. manufactured products	Dollar	0.02
Profit from production activities	Dollar	106.61
Profitability of production	%	11.7
The number of industrial and production personnel	persons	10
Productivity	Dollar/person	10.66

The production of 200kg low-fat chicken sausages per shift can make a profit of 106.61 Dollar, and the production profitability has reached 11.7%, indicating that the industrial production of low-fat chicken sausages with Ab has good economic prospect, huge benefits, and is worth promoting.

CONCLUSION OF SECTION 5

The economic benefit of low-fat chicken sausage production was analysed. The industrial production of low-fat chicken sausages with Ab has good economic prospect, huge benefits, and is worth promoting.

The production of 200kg low-fat chicken sausages per shift can make a profit of 106.61 Dollar, and the production profitability has reached 11.7%, indicating that the industrial production of low-fat chicken sausages with Ab has good economic prospect, huge benefits, and is worth promoting.

CONCLUSION

1. Based on previous studies, this study systematically analyzed the development trend of meat and meat products, the development status and trend of low-fat meat products, the characteristics and application of mushrooms in food, and the application prospect of Ab mushroom in low-fat meat products. The results are that chicken consumption will continue to grow in the long-term. Low-temperature meat products dominated the market of meat products. Future study on the creation of low-temperature meat products will focus heavily on low-fat meat products. Mushrooms are widely used in meat products and have broad application prospects in low-fat meat products.

2. Ab can act as a fat substitute to produce low-fat chicken products to meet the needs of consumers for nutritious and healthy meat products. At present, there is no study available yet on the application of Ab mushroom in low-fat chicken products. Therefore, Ab has a broad application prospect in low-fat chicken products.

3. Ab as a fat substitute affected the gel properties and microstructure of chicken batters. Under special substitution percentages, adding Ab mushroom significantly ($P < 0.05$) increased the pH value, and improved the emulsion stability, gel characteristics and microstructure of chicken batters, while the L* and whiteness of chicken batters with Ab powder decreased. In conclusion, Ab mushroom can act as a fat substitute to produce low-fat healthy chicken products. It was advised to replace 20% of pork back fat with Ab mushroom to obtain the best gel quality.

4. When Ab mushrooms alone or in combination with soybean oil or water as fat substitutes, the cooking yield, water holding capacity, texture, and rheological properties of the chicken batter were improved and a part of free water was transformed into immobilized water. However, the color of the chicken batter changed significantly ($P < 0.05$). Particularly, the combination of Ab mushroom and soybean oil demonstrated the optimal effect to improve the gel properties, rheology, microstructure and water-binding ability of the chicken batter. Thus, the results suggest that the concomitant use of Ab mushroom and soybean oil is a promising strategy for developing healthier chicken products.

5. When the compound of Ab and soybean oil was used as a fat substitute in chicken batters, its particle size should be considered. Large particle size was more conducive to the increase in pH value, cooking yield, and water holding capacity of chicken batters by converting a part of free water into bonding water. Small particle size was more conducive to the L* value, b* value, whiteness value and viscoelasticity of chicken batters, and the improvement of the microstructure of chicken batters.

6. Ab mushroom significantly affected the WHC, gel strength, texture, rheology, water distribution, microstructure, and protein secondary structure of chicken MP gels. Adding Ab mushroom increased the viscoelasticity of MP gels, caused a part of free water in MP gels to turn into immobilized water, and enhanced the WHC and gel strength of MP gels. Ab mushroom powder as a filler could enter the network matrix of the protein, forming a gel with smaller pores and greater density. A specific amount of Ab powder was beneficial to the texture of MP gels. Moreover, the interaction between Ab and protein resulted in the addition of β -folding and the reduction of α -helix during protein denaturation. In conclusion, MP gels with a denser and more uniform structure can be obtained when a specific amount of Ab mushroom is incorporated into MP, which has high water retention and improved texture. The study suggested that Ab could be a promising ingredient in improving chicken MP's gel properties and developing fat-reduced meat products.

7. The addition of potato starch, chicken essence, refined salt, and premix of Ab and soybean oil was optimized, and a low-fat, low-calorie, sodium nitrite-free healthy chicken sausage containing Ab was successfully developed. Compared with high-fat chicken sausages, low-fat chicken sausages had significantly lower energy and higher ash and carbohydrate content ($P < 0.05$). There were no significant changes in moisture, protein, total essential amino acids, and non-essential amino acids ($P < 0.05$). The contents of fat, SFA and MSFA were significantly reduced, and PSFA content increased by 2.5 times ($P < 0.05$). The cooking yield was close to 100%, hardness and chewiness increased significantly, springiness and cohesiveness did not change significantly, L* value decreased, and the values of a*, b*, pH and TBARS increased significantly ($P < 0.05$). Consumers enjoyed the flavor, texture, and overall acceptability of the Ab-

containing low-fat chicken sausage, and accepted its color and taste.

8. During the storage period of 35 days, the redness value of low-fat chicken sausage gradually increased, and the yellowness value gradually decreased, which showed a completely opposite trend of color change with high-fat chicken sausage. There were no changes in pH and cohesiveness, a fluctuation in chewiness and firmness, and a slight decrease in elasticity. Compared with high-fat chicken sausage, low-fat chicken sausage showed stronger antioxidant properties. Although the total bacterial count of low-fat chicken sausage was higher than that of high-fat chicken sausage, the increase of total bacterial count during storage was lower than that of high-fat chicken sausage. Low-fat chicken sausages have a minimum shelf life of 25 days.

9. The recipe developed for low fat chicken sausage is 60 grams of chicken breast, 8 grams of pork back fat, 4 grams of Ab powder, 8 grams of soybean oil, 20 grams of ice water, 1.4 grams of refined salt, and 0.3 grams of tripolyphosphate , 0.1 grams of chicken essence, 0.65 grams of sugar, 0.15 grams of white pepper, 5 grams of potato starch. The energy value of the developed low-fat chicken sausage containing Ab is 220.44kcal/100g. The nutrient contents are as follows: moisture 61.85%, ash 1.54%, protein 12.51%, fat 14.8%, carbohydrate 9.3%.

10. According to the best technological scheme obtained in this study, the economic benefits of the production of low-fat chicken sausages containing Ab mushroom were analyzed. It was found that the industrial production of low-fat chicken sausages has good economic prospects and huge benefits, which is worth promoting.

11. The production of 200kg low-fat chicken sausages per shift can make a profit of 106.6 Dollar, and the production profitability has reached 11.7%, indicating that the industrial production of low-fat chicken sausages with Ab has good economic prospect, huge benefits, and is worth promoting.

REFERENCES

[1] Higgs JD. The changing nature of red meat: 20 years of improving nutritional quality. Trends in Food Science & Technology. 2000;11（3）85-95.

[2] Yuefeng Y. China food composition table Beijing. 2019;6.

[3] FAO/WHO. Protein quality evaluation.pdf. 1991.

[4] Pereira PM, Vicente AF. Meat nutritional composition and nutritive role in the human diet. Meat science. 2013;93:586-92.

[5] Krauss RM, Eckel RH, Howard B, Appel LJ, Daniels SR, Deckelbaum RJ, et al. AHA Dietary Guidelines: revision 2000: A statement for healthcare professionals from the Nutrition Committee of the American Heart Association. Circulation. 2000;102:2284-99.

[6] Diallo A, Deschasaux M, Latino-Martel P, Hercberg S, Galan P, Fassier P, et al. Red and processed meat intake and cancer risk: Results from the prospective NutriNet-Santé cohort study. International Journal of Cancer. 2018;142:230-7.

[7] Gordon A, Barbut S. Mechanisms of meat batter stabilization: a review. Critical reviews in food science and nutrition. 1992;32:299-332.

[8] Tornberg E. Effects of heat on meat proteins - Implications on structure and quality of meat products. Meat science. 2005;70:493-508.

[9] Kim T-K, Hwang K-E, Sung J-M, Park J-D, Kim M-H, Jeon K-H, et al. Replacement of pork back fat with pre-emulsion of wheat (Triticum aestivumL.) sprout and collagen and its optimization for reduced-fat patties. Journal of Food Processing and Preservation. 2018;42:e13576.

[10] Brewer MS. Reducing the fat content in ground beef without sacrificing quality: a review. Meat science. 2012;91:385-95.

[11] Kumar Y. Development of Low-Fat/Reduced-Fat Processed Meat Products using Fat Replacers and Analogues. Food Reviews International. 2019;37:296-312.

[12] Salles C, Chagnon MC, Feron G, Guichard E, Laboure H, Morzel M, et al. In-mouth mechanisms leading to flavor release and perception. Critical reviews in food science and nutrition. 2011;51:67-90.

[13] Khan MI, Jo C, Tariq MR. Meat flavor precursors and factors influencing flavor

precursors--A systematic review. Meat science. 2015;110:278-84.

[14] Brewer S, Novakofski J. Consumer sensory evaluations of aging effects on beef quality. Journal of food science. 2008;73:S78-82.

[15] Fiorentini M, Kinchla AJ, Nolden AA. Role of Sensory Evaluation in Consumer Acceptance of Plant-Based Meat Analogs and Meat Extenders: A Scoping Review. Foods. 2020;9.

[16] Shahidi F. Flavor of meat, meat products, and seafoods. International Thomson Publishing. 1998.

[17] Wu M, Xiong YL, Chen J. Rheology and microstructure of myofibrillar protein–plant lipid composite gels: Effect of emulsion droplet size and membrane type. Journal of Food Engineering. 2011;106:318-24.

[18] Wu M, Xiong YL, Chen J, Tang X, Zhou G. Rheological and microstructural properties of porcine myofibrillar protein-lipid emulsion composite gels. Journal of food science. 2009;74:E207-17.

[19] Varga-Visi É, Toxanbayeva B. Application of fat replacers and their effect on quality of comminuted meat products with low lipid content: A review. Acta Alimentaria. 2017;46:181-6.

[20] Arena R, Guazzi M, Lianov L, Whitsel L, Berra K, Lavie CJ, et al. Healthy Lifestyle Interventions to Combat Noncommunicable Disease-A Novel Nonhierarchical Connectivity Model for Key Stakeholders: A Policy Statement From the American Heart Association, European Society of Cardiology, European Association for Cardiovascular Prevention and Rehabilitation, and American College of Preventive Medicine. Mayo Clinic proceedings. 2015;90:1082-103.

[21] L.O. Fiems SDC, S. De Smet, G. Van de Voorde, J.M. Vanacker, Ch. V. Boucque . Relationship between fat depots in carcasses of beef bulls and effect on meat color and tenderness. Meat science. 2000;56:41-7.

[22] Kurcubic V, Okanovic D, Vasilev D, Ivic M, Colovic D, Jokanovic M, et al. Effects of replacing pork back fat with cellulose fiber in Pariser sausages. Fleischwirtschaft. 2020;100:82-8.

[23] Sorapukdee S, Jansa S, Tangwatcharin P. Partial replacement of pork back fat with

konjac gel in Northeastern Thai fermented sausage (Sai Krok E-san). Asian-Australasian journal of animal sciences. 2019;32:1763-75.

[24] Supavititpatana T, Apichartsrangkoon A. Combination effects of ultra-high pressure and temperature on the physical and thermal properties of ostrich meat sausage (yor). Meat science. 2007;76:555-60.

[25] Zhou F, Dong H, Shao JH, Zhang JL, Liu DY. Effect of chopping time and heating on (1) H nuclear magnetic resonance and rheological behavior of meat batter matrix. Animal science journal = Nihon chikusan Gakkaiho. 2018;89:695-702.

[26] Alarcon-Rojo AD, Carrillo-Lopez LM, Reyes-Villagrana R, Huerta-Jimenez M, Garcia-Galicia IA. Ultrasound and meat quality: A review. Ultrasonics sonochemistry. 2019;55:369-82.

[27] Cummings JH, Stephen AM. Carbohydrate terminology and classification. European journal of clinical nutrition. 2007;61 Suppl 1:S5-18.

[28] Jiménez-Colmenero F, Cofrades S, Herrero AM, Fernández-Martín F, Rodríguez-Salas L, Ruiz-Capillas C. Konjac gel fat analogue for use in meat products: Comparison with pork fats. Food Hydrocolloids. 2012;26:63-72.

[29] Marchetti L, Andres SC, Califano AN. Low-fat meat sausages with fish oil: optimization of milk proteins and carrageenan contents using response surface methodology. Meat science. 2014;96:1297-303.

[30] Akesowan A. Production and storage stability of formulated chicken nuggets using konjac flour and shiitake mushrooms. Journal of food science and technology. 2016;53:3661-74.

[31] Choe J, Kim HY. Quality characteristics of reduced fat emulsion-type chicken sausages using chicken skin and wheat fiber mixture as fat replacer. Poultry science. 2019;98:2662-9.

[32] Song J, Pan T, Wu J, Ren F. The improvement effect and mechanism of citrus fiber on the water-binding ability of low-fat frankfurters. Journal of food science and technology. 2016;53:4197-204.

[33] Zhuang X, Han M, Kang ZL, Wang K, Bai Y, Xu XL, et al. Effects of the sugarcane dietary fiber and pre-emulsified sesame oil on low-fat meat batter physicochemical

property, texture, and microstructure. Meat science. 2016;113:107-15.

[34] Zhuang X, Jiang X, Han M, Kang Z-l, Zhao L, Xu X-l, et al. Influence of sugarcane dietary fiber on water states and microstructure of myofibrillar protein gels. Food Hydrocolloids. 2016;57:253-61.

[35] Zhuang X, Zhang W, Liu R, Liu Y, Xing L, Han M, et al. Improved gel functionality of myofibrillar proteins incorporation with sugarcane dietary fiber. Food Res Int. 2017;100:586-94.

[36] Ferjančič B, Kugler S, Korošec M, Polak T, Bertoncelj J. Development of low-fat chicken bologna sausages enriched with inulin, oat fibre or psyllium. International Journal of Food Science & Technology. 2020;56:1818-28.

[37] Zhao Y, Hou Q, Zhuang X, Wang Y, Zhou G, Zhang W. Effect of regenerated cellulose fiber on the physicochemical properties and sensory characteristics of fat-reduced emulsified sausage. Lwt. 2018;97:157-63.

[38] Zeng H, Chen J, Zhai J, Wang H, Xia W, Xiong YL. Reduction of the fat content of battered and breaded fish balls during deep-fat frying using fermented bamboo shoot dietary fiber. Lwt. 2016;73:425-31.

[39] Henning SSC, Tshalibe P, Hoffman LC. Physico-chemical properties of reduced-fat beef species sausage with pork back fat replaced by pineapple dietary fibres and water. Lwt. 2016;74:92-8.

[40] Zhao D, Guo C, Liu X, Xiao C. Effects of insoluble dietary fiber from kiwi fruit pomace on the physicochemical properties and sensory characteristics of low-fat pork meatballs. Journal of food science and technology. 2021;58:1524-37.

[41] Marchetti L, Muzzio B, Cerrutti P, Andrés SC, Califano AN. Bacterial nanocellulose as novel additive in low-lipid low-sodium meat sausages. Effect on quality and stability. Food Structure. 2017;14:52-9.

[42] Paglarini CS, Vidal VAS, Ozaki MM, Ribeiro APB, Bernardinelli OD, Camara A, et al. Inulin gelled emulsion as a fat replacer and fiber carrier in healthier Bologna sausage. Food science and technology international = Ciencia y tecnologia de los alimentos internacional. 2022;28:3-14.

[43] Silva Moraes K, Rafael R, Salas-Mellado M, Prentice C. Influence of chicken protein

isolate and heating temperature on textural properties of low-fat frankfurters. Journal of Applied Poultry Research. 2011;20:253-62.

[44] Malav OP, Talukder S, Gokulakrishnan P, Chand S. Meat analog: a review. Critical reviews in food science and nutrition. 2015;55:1241-5.

[45] Asgar MA, Fazilah A, Huda N, Bhat R, Karim AA. Nonmeat Protein Alternatives as Meat Extenders and Meat Analogs. Comprehensive Reviews in Food Science and Food Safety. 2010;9:513-29.

[46] Chen Y, Jia X, Sun F, Jiang S, Liu H, Liu Q, et al. Using a stable pre-emulsified canola oil system that includes porcine plasma protein hydrolysates and oxidized tannic acid to partially replace pork fat in frankfurters. Meat science. 2020;160:107968.

[47] Choi YS, Choi JH, Han DJ, Kim HY, Lee MA, Jeong JY, et al. Effects of replacing pork back fat with vegetable oils and rice bran fiber on the quality of reduced-fat frankfurters. Meat science. 2010;84:557-63.

[48] De Carvalho FAL, Munekata PES, Pateiro M, Campagnol PCB, Domínguez R, Trindade MA, et al. Effect of replacing back fat with vegetable oils during the shelf-life of cooked lamb sausages. Lwt. 2020;122:109052.

[49] Zhu X, Li L, Li S, Ning C, Zhou C. l–Arginine/l–lysine improves emulsion stability of chicken sausage by increasing electrostatic repulsion of emulsion droplet and decreasing the interfacial tension of soybean oil-water. Food Hydrocolloids. 2019;89:492-502.

[50] Wang X, Xie Y, Li X, Liu Y, Yan W. Effects of partial replacement of pork back fat by a camellia oil gel on certain quality characteristics of a cooked style Harbin sausage. Meat science. 2018;146:154-9.

[51] Tan SS, Aminah A, Zhang XG, Abdul SB. Optimizing palm oil and palm stearin utilization for sensory and textural properties of chicken frankfurters. Meat science. 2006;72:387-97.

[52] Kaynakci E, KiliÇ B. Effect of replacing beef fat with safflower oil on physicochemical, nutritional and oxidative stability characteristics of wieners. Food Science and Technology. 2021;41:52-9.

[53] Rodriguez-Carpena JG, Morcuende D, Estevez M. Avocado, sunflower and olive oils as replacers of pork back-fat in burger patties: effect on lipid composition, oxidative stability

and quality traits. Meat science. 2012;90:106-15.

[54] Nieto G, Lorenzo JM. Use of olive oil as fat replacer in meat emulsions. Current Opinion in Food Science. 2021;40:179-86.

[55] Ferro AC, de Souza Paglarini C, Rodrigues Pollonio MA, Lopes Cunha R. Glyceryl monostearate-based oleogels as a new fat substitute in meat emulsion. Meat science. 2021;174:108424.

[56] Isabel C.F.R. Ferreira ÂFaSAH. Chemical, Nutritional, and Bioactive Potential of Mushrooms. Wiley. 2017;8:455-501.

[57] Daniel J. Royse JBaQT. Current Overview of Mushroom Production in the World. Edible and medicinal

mushrooms: technology and applications John Wiley & Sons LtD, Hoboken,. 2017;8:5-13.

[58] Food and Agriculture Organization(2020). FAOSTAT – Crop. Retrieved January 30. from http://wwwfaoorg/faostat/en/#data/Q.

[59] China Edible Fungi Association. Analysis of statistical survey results of edible fungi in China in 2021. Beijing. 2022.12.

[60] Chang ST, Wasser SP. The Role of Culinary-Medicinal Mushrooms on Human Welfare with a Pyramid Model for Human Health. International Journal of Medicinal Mushrooms. 2012;14:95-134.

[61] Hans-Peter Hanssen AK. Determination of some Important Flavor Compounds in Commercial Mushroom Concentrates. Z Lebensm Unters Forsch 1983;177:333-5.

[62] Correa RC, de Souza AH, Calhelha RC, Barros L, Glamoclija J, Sokovic M, et al. Bioactive formulations prepared from fruiting bodies and submerged culture mycelia of the Brazilian edible mushroom Pleurotus ostreatoroseus Singer. Food & function. 2015;6:2155-64.

[63] Muszynska B, Grzywacz-Kisielewska A, Kala K, Gdula-Argasinska J. Anti-inflammatory properties of edible mushrooms: A review. Food chemistry. 2018;243:373-81.

[64] Kalaras MD, Richie JP, Calcagnotto A, Beelman RB. Mushrooms: A rich source of the antioxidants ergothioneine and glutathione. Food chemistry. 2017;233:429-33.

[65] Szabó A, Geösel A, Kókai Z, Orbán C, Töreki K, Szőke A. Antioxidant activity as

indicator of UV radiation and other abiotic stress factors on Agaricus bisporus (Lange/Imbach) and Sedum hybridum (L.). Acta Universitatis Sapientiae, Alimentaria. 2016;9:90-100.

[66] Khan AA, Gani A, Khanday FA, Masoodi FA. Biological and pharmaceutical activities of mushroom β-glucan discussed as a potential functional food ingredient. Bioactive Carbohydrates and Dietary Fibre. 2018;16:1-13.

[67] Kang MY, Rico CW, Lee SC. In vitro antioxidative and antimutagenic activities of oak mushroom (Lentinus edodes) and king oyster mushroom (Pleurotus eryngii) byproducts. Food Science and Biotechnology. 2012;21:167-73.

[68] Nayak PC, Raju CV, Lakshmisha IP, Singh RR, Sofi FR. Influence of Button mushroom (Agaricus bisporus) on quality and refrigerated storage stability of patties prepared from sutchi catfish (Pangasius hypophthalmus). Journal of food science and technology. 2015;52:3529-38.

[69] Li B, Lu F, Suo X, Nan H, Li B. Antioxidant properties of cap and stipe from Coprinus comatus. Molecules. 2010;15:1473-86.

[70] Bo Li Fl, Xiao-Min Suo. Glucose lowering activity of Coprinus comatus. Agrofood industry hi-tech. 2010;21:15-7.

[71] Van Doan H, Doolgindachbaporn S, Suksri A. Effects of Eryngii mushroom (Pleurotus eryngii) and Lactobacillus plantarum on growth performance, immunity and disease resistance of Pangasius catfish (Pangasius bocourti, Sauvage 1880). Fish physiology and biochemistry. 2016;42:1427-40.

[72] Guillamon E, Garcia-Lafuente A, Lozano M, D'Arrigo M, Rostagno MA, Villares A, et al. Edible mushrooms: role in the prevention of cardiovascular diseases. Fitoterapia. 2010;81:715-23.

[73] Ren Z, Li J, Song X, Zhang J, Wang W, Wang X, et al. The regulation of inflammation and oxidative status against lung injury of residue polysaccharides by Lentinula edodes. International journal of biological macromolecules. 2018;106:185-92.

[74] Gunawardena D, Shanmugam K, Low M, Bennett L, Govindaraghavan S, Head R, et al. Determination of anti-inflammatory activities of standardised preparations of plant- and mushroom-based foods. European journal of nutrition. 2014;53:335-43.

[75] Komura DL, Carbonero ER, Gracher AH, Baggio CH, Freitas CS, Marcon R, et al. Structure of Agaricus spp. fucogalactans and their anti-inflammatory and antinociceptive properties. Bioresource technology. 2010;101:6192-9.

[76] Doymaz İ. Infrared drying of button mushroom slices. Food Science and Biotechnology. 2014;23:723-9.

[77] Bhattacharya M, Srivastav PP, Mishra HN. Thin-layer modeling of convective and microwave-convective drying of oyster mushroom (Pleurotus ostreatus). Journal of food science and technology. 2015;52:2013-22.

[78] Kantrong H, Tansakul A, Mittal GS. Drying characteristics and quality of shiitake mushroom undergoing microwave-vacuum drying and microwave-vacuum combined with infrared drying. Journal of food science and technology. 2014;51:3594-608.

[79] Giavasis I. Bioactive fungal polysaccharides as potential functional ingredients in food and nutraceuticals. Current opinion in biotechnology. 2014;26:162-73.

[80] Zhu F, Du B, Bian Z, Xu B. Beta-glucans from edible and medicinal mushrooms: Characteristics, physicochemical and biological activities. Journal of Food Composition and Analysis. 2015;41:165-73.

[81] Fan J, Zhang J, Tang Q, Liu Y, Zhang A, Pan Y. Structural elucidation of a neutral fucogalactan from the mycelium of Coprinus comatus. Carbohydrate research. 2006;341:1130-4.

[82] Andriy Synytsya Km, Ivan Jablonský, Marcela Sluková, Jana Čopíková. Mushrooms of Genus Pleurotus as a Source of Dietary Fibres. Czech J Food Sci. 2008;26:441-6

[83] Wenhang Wang CL, Guanhua Du, Xiuling Zhang, and Hongjie Zhang. Characteristics and Rheological Properties of Polysaccharide Nanoparticles from Edible Mushrooms (Flammulina velutipes). Journal of food science. 2017;82:687-93.

[84] Andriy Synytsya Km, Ivan Jablonský, Marcela Sluková, Jana Čopíková. Mushrooms of Genus Pleurotus as a Source of Dietary Fibres and Glucans for Food Supplements. Czech J Food Sci. 2008;26:441–6.

[85] Zhaojing Wang DL, Zhongyan Liang. Structure of polysaccharides from the fruiting body of Hericium erinaceus Pers. Carbohydrate polymers. 2004;57:241-7.

[86] Mondal S, Chakraborty I, Pramanik M, Rout D, Islam SS. Structural studies of water-

soluble polysaccharides of an edible mushroom, Termitomyces eurhizus. A reinvestigation. Carbohydrate research. 2004;339:1135-40.

[87] Li. B, Dobruchowska JM, Gerwig GJ, Dijkhuizen L, Kamerling JP. Structural investigation of water-soluble polysaccharides extracted from the fruit bodies of Coprinus comatus. Carbohydrate polymers. 2013;91:314-21.

[88] Heo S, Lee SM, Bae IY, Park H-G, Lee HG, Lee S. Effect of Lentinus edodes β-Glucan-Enriched Materials on the Textural, Rheological, and Oil-Resisting Properties of Instant Fried Noodles. Food and Bioprocess Technology. 2011;6:553-60.

[89] ZHANG Mengtian YW, PEI Fei, ZHAO Liyan, AN Xinxin, MA Ning, CHENG Wei, HU Qiuhui. Optimization of Enzymatic Hydrolysis for Preparing Pleurotus eryngii Protein by Response Surface Methodology
and Nutritional Evaluation. Food Science. 2015;36:125-30.

[90] Haverstock K, Forgays DK. To eat or not to eat. A comparison of current and former animal product limiters. Appetite. 2012;58:1030-6.

[91] Suwimol Chockchaisawasdee SN, Singdong Pochana and Costas E. Stathopoulos. Development of fermented oyster-mushroom sausage. As J Food Ag-Ind. 2010;3:35-43.

[92] Emy Yuliantini K. The Formulation Of Analog Sausage With High Protein And Cork Sea Fiber (Channa Striata), And Oyster Mushroom As The Healthy Snacks For The School Children. Advances in Health Sciences Research (AHSR). 2019;14:192-203.

[93] Nagy M, Semeniuc CA, Socaci SA, Pop CR, Rotar AM, SĂLĂGean CD, et al. Utilization of brewer's spent grain and mushrooms in fortification of smoked sausages. Food Science and Technology. 2017;37:315-20.

[94] Stepanova T, Kondratjuk N, Haijuan N, Suprunenko K. Prospects of cultivated mushrooms use in technology of sausages. Bulletin of the National Technical University «KhPI» Series: New solutions in modern technologies. 2019;0:75-80.

[95] Wang X, Zhou P, Cheng J, Chen Z, Liu X. Use of straw mushrooms (Volvariella volvacea) for the enhancement of physicochemical, nutritional and sensory profiles of Cantonese sausages. Meat science. 2018;146:18-25.

[96] Wang L, Guo H, Liu X, Jiang G, Li C, Li X, et al. Roles of Lentinula edodes as the pork lean meat replacer in production of the sausage. Meat science. 2019;156:44-51.

[97] Kyung Jo JL, Samooel Jung. Quality Characteristics of Low-salt Chicken Sausage Supplemented with a Winter Mushroom Powder. Korean journal for food science of animal resources. 2018;38:768-79.

[98] Flávia Santi Stefanello, Mariana dos Santos da Silva, CPC, Leadir Lucy Martins Fries, , Fernanda Luísa Ludtke, Ernesto Hashime Kubota. Oxidative and microbiological stability of fresh pork sausage with added sun mushroom powder. Ciênc Agrotec, Lavras. 2015;39:381-9.

[99] Stefanello FS, Cavalheiro CP, Ludtke FL, Silva MDSd, Fries LLM, Kubota EH. Effect of sun mushroom extract in pork sausage and evaluation of the oxidative and microbiological stability of the product. Semina: Ciências Agrárias. 2015;36:171-85.

[100] LV Deping WT, CHEN Xu. Research on the Development of Edible Fungi Drinks. Edible fungi of China. 2013;32:1-3.

[101] Li L. Advances on mushroom compounded functional drink. Food Science and Technology. 2013;38:110-4.

[102] Zhou Jiaqian Wx, Shao Juanjuan,Wang Linlin. Research Progress of Fermented Beverage of Edible Fungi. Farm Products Processing. 2017:60-3.

[103] Feng T, Wu Y, Zhang Z, Song S, Zhuang H, Xu Z, et al. Purification, Identification, and Sensory Evaluation of Kokumi Peptides from Agaricus bisporus Mushroom. Foods. 2019;43:1-12.

[104] Shen Wenfeng WY. Research Status on Process Technology of Edible Fungi Seasoning. Farm Products Processing. 2016:65-7.

[105] SHEN Wenfeng WW, CUI Wenjia, SHI Xianquan. Research Status and Development Trends of Edible Fungi Seasoning. Food and Nutrition in China. 2016;22:35-8.

[106] Liu Dayu ZL, Xiao Longquan, Li Zongtang, Wang Junding. Processing and preservation technology of compound seasoning of Edible fungus and bones. China condiment. 2015;40:81-3.

[107] YIN Ling CS, ZHAO Liyan, PEI Fei, YANG Wenjian, Hu Qiuhui. Quality Characteristics of Straw Mushrooms (Volvariella volvacea) Chips Prepared by Low-Temperature Vacuum Frying and Vacuum Freeze Drying. Food Science. 2017;38:192-9.

[108] Kumar K, Barman Ray A. Development and shelf-life evaluation of tomato-mushroom mixed ketchup. Journal of food science and technology. 2016;53:2236-43.

[109] Petrović J, Glamočlija J, Stojković D, Ćirić A, Barros L, Ferreira ICFR, et al. Nutritional value, chemical composition, antioxidant activity and enrichment of cream cheese with chestnut mushroom Agrocybe aegerita (Brig.) Sing. Journal of food science and technology. 2015;52:6711-8.

[110] Liyan GYHQ-hYWFYMNZ. Production of Lentinus edodes Biscuits and Analysis of Characteristic Aroma Components. Food Science. 2013;34:58-63.

[111] YANG Wenjian YJ, SUN Yong, MA Ning, FANG Yong, PEI Fei, HU Qiuhui, XIANG Chunrong. Effect of Flammulina velutipes and Agrocybe aegirit Powders on Rheological Properties of Doug. Food Science. 2014;35:43-7.

[112] CHANG Shijie YZ, YIN Ling, HU Qiuhui, PEI Fei, YANG Wenjian, ZHAO Liyan. Processing of Spicy Volvaria volvacea and Its Flavor Characteristics. Food Science. 2018;39:135-40.

[113] Yuan B, Zhao L, Yang W, McClements DJ, Hu Q. Enrichment of Bread with Nutraceutical-Rich Mushrooms: Impact of Auricularia auricula (Mushroom) Flour Upon Quality Attributes of Wheat Dough and Bread. Journal of food science. 2017;82:2041-50.

[114] Devina Vaidya GS, Rai RD, Sharma PC. Development and quality evaluation of white button mushroom noodles. J Food Sci Technol. 2008; 45 513-5.

[115] Se-Young Kim S-IC, Seok-Hyun Nam, Mi-Young Kang. Cholesterol Lowering Action and Antioxidant Status Improving Efficacy of Noodles Made from Unmarketable Oak Mushroom (Lentinus edodes) in High Cholesterol Fed Rats. Journal of the Korean Society for Applied Biological Chemistry. 2009;52:207-12.

[116] Chung SI, Kim SY, Nam YJ, Kang MY. Development of surimi gel from king oyster mushroom and cuttlefish meat paste. Food Science and Biotechnology. 2010;19:51-6.

[117] Sharma A, Vaidya D, Abrol GS, Rana N, Chauhan N. Functional and textural properties of Indian nuggets assorted with mushroom for lysine enrichment. Journal of food science and technology. 2015;52:3837-42.

[118] Usman M, Murtaza G, Ditta A. Nutritional, Medicinal, and Cosmetic Value of Bioactive Compounds in Button Mushroom (Agaricus bisporus): A Review. Applied Sciences.

2021;11:5943.

[119] Bilal AW, R HB, A HW. Nutritional and medicinal importance of mushrooms. Journal of Medicinal Plants Research. 2010;4:2598-604.

[120] Apoorva Bhushan, Kulshreshtha M. The Medicinal Mushroom Agaricus bisporus: Review of Phytopharmacology and Potential Role in the Treatment of Various Diseases. Journal of Nature and Science of Medicine. 2018;1:4-9.

[121] SUNDAR RAO KOYYALAMUDI, JEONG S-C, CHI-HYUN SONG, KAI YIP CHO, PANG G. Vitamin D2 formation and bioavailability from Agaricus bisporus button mushrooms treated with ultraviolet irradiation. Journal of agricultural and food chemistry. 2009;57:3351-5.

[122] MATTILA，P, PIRJO SALO-VAANANEN, NKO，KK, HEIKKI ARO, JALAVA T. Basic Composition and Amino Acid Contents of Mushrooms Cultivated in Finland. J Agric Food Chem. 2002;50:6149-422.

[123] Sánchez C. Bioactives from Mushroom and Their Application. M Puri (ed), Food Bioactives. 2017:23-57.

[124] Ozturk M, Duru ME, Kivrak S, Mercan-Dogan N, Turkoglu A, Ozler MA. In vitro antioxidant, anticholinesterase and antimicrobial activity studies on three Agaricus species with fatty acid compositions and iron contents: a comparative study on the three most edible mushrooms. Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association. 2011;49:1353-60.

[125] Blumfield M, Abbott K, Duve E, Cassettari T, Marshall S, Fayet-Moore F. Examining the health effects and bioactive components in Agaricus bisporus mushrooms: a scoping review. Journal Of Nutritional Biochemistry. 2020;84.

[126] Ting L. Structural improvement of China's meat industry chain. Hongta Securities Co, Ltd Research and Development Center. 2018.

[127] Kang Z-L, Zhang X-h, Li X, Song Z-j, Ma H-j, Lu F, et al. The effects of sodium chloride on proteins aggregation, conformation and gel properties of pork myofibrillar protein Running Head: Relationship aggregation, conformation and gel properties. Journal of food science and technology. 2020;25:1-7.

[128] Gornall AG, Bardawill CJ, David MM. Determination Of Serum Proteins by Means Of the Biuret Reaction. Journal of Biological Chemistry. 1949;177:751-66.

[129] Choe J, Lee J, Jo K, Jo C, Song M, Jung S. Application of winter mushroom powder as an alternative to phosphates in emulsion-type sausages. Meat science. 2018;143:114-8.

[130] Fernandez-Martin F, Lopez-Lopez I, Cofrades S, Colmenero FJ. Influence of adding Sea Spaghetti seaweed and replacing the animal fat with olive oil or a konjac gel on pork meat batter gelation. Potential protein/alginate association. Meat science. 2009;83:209-17.

[131] Wang L, Li C, Ren L, Guo H, Li Y. Production of Pork Sausages Using Pleaurotus eryngii with Different Treatments as Replacements for Pork Back Fat. Journal of food science. 2019;84:3091-8.

[132] Wattanachant S, Benjakul S, Ledward DA. Effect of heat treatment on changes in texture, structure and properties of Thai indigenous chicken muscle. Food chemistry. 2005;93:337-48.

[133] Liu F, Dong X, Shen S, Shi Y, Ou Y, Cai W, et al. Changes in the digestion properties and protein conformation of sturgeon myofibrillar protein treated by low temperature vacuum heating during in vitro digestion. Food & function. 2021;12:6981-91.

[134] AOAC. Official method of analysis. Maryland, USA: Association of Official Analytical Chemistry. 2005.

[135] Carvalho Barros J, Munekata PES, de Carvalho FAL, Pateiro M, Barba FJ, Dominguez R, et al. Use of Tiger Nut (Cyperus esculentus L.) Oil Emulsion as Animal Fat Replacement in Beef Burgers. Foods. 2020;9.

[136] Xu D, Zhang H, Zhang L, Huang T, Jia R, Lu J, et al. Effects of electron beam irradiation on quality of weever fillets during refrigerated storage. Food Frontiers. 2021;2:519-26.

[137] National food safety standard of China. Microbiological examination of food-Determination of total bacterial count(2016).GB 4789.2-2016.

[138] Ito H, Ueno H, Kikuzaki H. Construction of a free-form amino acid database for vegetables and mushrooms. Integrative Food, Nutrition and Metabolism. 2017;4:1-9.

[139] Kurt A, Gençcelep H. Enrichment of meat emulsion with mushroom (Agaricus

bisporus) powder: Impact on rheological and structural characteristics. Journal of Food Engineering. 2018;237:128-36.

[140] Barbut S. Microstructure of white and dark turkey meat batters as affected by pH. British poultry science. 1997;38:175-82.

[141] Lee J, Jo K, Lim Y, Jeon HJ, Choe JH, Jo C, et al. The use of atmospheric pressure plasma as a curing process for canned ground ham. Food chemistry. 2018;240:430-6.

[142] Han M, Bertram HC. Designing healthier comminuted meat products: Effect of dietary fibers on water distribution and texture of a fat-reduced meat model system. Meat science. 2017;133:159-65.

[143] Chaplin MF. Fibre and water binding. Proceedings of the Nutrition Society. 2007;62:223-7.

[144] Ha J-U, Koo S-G, Lee H-Y, Hwang Y-M, Lee S-C. Physical Properties of fish Paste Containing Agaricus bisporus. Korean Journal of Food Science and Technology. 2001;33:451-4.

[145] Wong KM, Decker EA, Autio WR, Toong K, DiStefano G, Kinchla AJ. Utilizing Mushrooms to Reduce Overall Sodium in Taco Filling Using Physical and Sensory Evaluation. Journal of food science. 2017;82:2379-86.

[146] Hong-Sun, Yook, Ju-Woon, Lee, Hyun-Ja, Lee, et al. Quality Properties of Fish Paste Prepared with Refined Dietary Fiber from Ascidian (Halocynthia roretzi) Tunic. Journal of the Korean Society of Food Science and Nutrition. 2000;29:642-6.

[147] Barbut AGS. Effect of Chloride Salts on Protein Extraction and Interfacial Protein Film Formation in Meat Batters. J Sci Food Agric. 1992;58:227-38.

[148] Westphalen AD, Briggs JL, Lonergan SM. Influence of muscle type on rheological properties of porcine myofibrillar protein during heat-induced gelation. Meat science. 2006;72:697-703.

[149] Mleczek M, Budka A, Siwulski M, Mleczek P, Gąsecka M, Jasińska A, et al. Investigation of differentiation of metal contents of Agaricus bisporus, Lentinula edodes and Pleurotus ostreatus sold commercially in Poland between 2009 and 2017. Journal of Food Composition and Analysis. 2020;90:1-9.

[150] Juqing HFCHWLGXZGW. Effect of sweet potato dietary fiber on water holding

capacity and gelling properties of myofibrillar proteins. Journal of Nanjing Agricultural University. 2019;42: 932 − 9

[151] Liu J, Lv Y, Mo X, Duan S, Tong Q. Effects of freezing and thawing treatment on the rheological and textural characteristics and micro-structure of heat-induced egg yolk gels. Journal of Food Engineering. 2018;216:144-50.

[152] Verma AK, Sharma BD, Banerjee R. Effect of sodium chloride replacement and apple pulp inclusion on the physico-chemical, textural and sensory properties of low fat chicken nuggets. LWT - Food Science and Technology. 2010;43:715-9.

[153] Cerón-Guevara MI, Rangel-Vargas E, Lorenzo JM, Bermúdez R, Pateiro M, Rodriguez JA, et al. Effect of the addition of edible mushroom flours (Agaricus bisporus and Pleurotus ostreatus) on physicochemical and sensory properties of cold-stored beef patties. Journal of Food Processing and Preservation. 2019;44:14351.

[154] Zhuang X, Han M, Bai Y, Liu Y, Xing L, Xu X-l, et al. Insight into the mechanism of myofibrillar protein gel improved by insoluble dietary fiber. Food Hydrocolloids. 2018;74:219-26.

[155] Zhao Y, Zhou G, Zhang W. Effects of regenerated cellulose fiber on the characteristics of myofibrillar protein gels. Carbohydrate polymers. 2019;209:276-81.

[156] Vidal VAS, Paglarini CdS, Ferreira A, Santos JRd, Pollonio MAR. Influence of the addition of KCl and CaCl2 blends on the physicochemical parameters of salted meat products throughout the processing steps. Food Science and Technology. 2020;40:665-70.

[157] Qing Z, Cheng J, Wang X, Tang D, Liu X, Zhu M. The effects of four edible mushrooms (Volvariella volvacea, Hypsizygus marmoreus, Pleurotus ostreatus and Agaricus bisporus) on physicochemical properties of beef paste. Lwt. 2021;135:110063.

[158] Jurić S, Jurić M, Król-Kilińska Ż, Vlahoviček-Kahlina K, Vinceković M, Dragović-Uzelac V, et al. Sources, stability, encapsulation and application of natural pigments in foods. Food Reviews International. 2020:1-56.

[159] Steglich MGW. Pigments of Fungi (Macromycetes). Progress in the Chemistry of Organic Natural Products. 1987:253-86.

[160] Zhang X, Wang W, Wang Y, Wang Y, Wang X, Gao G, et al. Effects of nanofiber

cellulose on functional properties of heat-induced chicken salt-soluble meat protein gel enhanced with microbial transglutaminase. Food Hydrocolloids. 2018;84:1-8.

[161] Choi YS, Choi JH, Han DJ, Kim HY, Lee MA, Kim HW, et al. Characteristics of low-fat meat emulsion systems with pork fat replaced by vegetable oils and rice bran fiber. Meat science. 2009;82:266-71.

[162] Sha L, Liu S, Liu D. Effects of soybean protein isolate on protein structure, batter rheology, and water migration in emulsified sausage. Journal of Food Processing and Preservation. 2020;44:1-10.

[163] Cando D, Herranz B, Borderías AJ, Moreno HM. Effect of high pressure on reduced sodium chloride surimi gels. Food Hydrocolloids. 2015;51:176-87.

[164] Câmara AKFI, Okuro PK, Cunha RLd, Herrero AM, Ruiz-Capillas C, Pollonio MAR. Chia (Salvia hispanica L.) mucilage as a new fat substitute in emulsified meat products: Technological, physicochemical, and rheological characterization. Lwt. 2020;125:109193.

[165] Sánchez-González I, Rodríguez-Casado A, Careche M, Carmona P. Raman analysis of surimi gelation by addition of wheat dietary fibre. Food chemistry. 2009;112:162-8.

[166] Pearce KL, Rosenvold K, Andersen HJ, Hopkins DL. Water distribution and mobility in meat during the conversion of muscle to meat and ageing and the impacts on fresh meat quality attributes--a review. Meat science. 2011;89:111-24.

[167] Luo H, Guo C, Lin L, Si Y, Gao X, Xu D, et al. Combined Use of Rheology, LF-NMR, and MRI for Characterizing the Gel Properties of Hairtail Surimi with Potato Starch. Food and Bioprocess Technology. 2020;13:637-47.

[168] Li W, Wang P, Xu X, Xing T, Zhou G. Use of low-field nuclear magnetic resonance to characterize water properties in frozen chicken breasts thawed under high pressure. European Food Research and Technology. 2014;239:183-8.

[169] Li K, Liu JY, Fu L, Zhao YY, Zhu H, Zhang YY, et al. Effect of bamboo shoot dietary fiber on gel properties, microstructure and water distribution of pork meat batters. Asian-Australasian journal of animal sciences. 2020;33:1180-90.

[170] McDonnell CK, Allen P, Duggan E, Arimi JM, Casey E, Duane G, et al. The effect of salt and fibre direction on water dynamics, distribution and mobility in pork muscle: a

low field NMR study. Meat science. 2013;95:51-8.

[171] Han M, Zhang Y, Fei Y, Xu X, Zhou G. Effect of microbial transglutaminase on NMR relaxometry and microstructure of pork myofibrillar protein gel. European Food Research and Technology. 2008;228:665-70.

[172] Trout GR. Techniques for Measuring Water-Binding Capacity in Muscle Foods A Review of Methodology. Meat science. 1988;23:235–52.

[173] SCHMIDT DGSGR. IONIC, pH， AND TEMPERATURE EFFECTS ON THE BINDING ABILITY OF MYOSIN. Journal of food science. 1979;44:1686-9.

[174] S. N. Heller LRH, J. M. Rivers, P. J. Van Soest, D. A. Roe, M.D., B. A. Lewis, J. Robertson Dietary fiber: the effect of particle size of wheat bran on colonic function in young adult men1. The American Journal of Clinical Nutrition. 1980;33:1734-44.

[175] Sangnark A, Noomhorm A. Chemical, physical and baking properties of dietary fiber prepared from rice straw. Food Research International. 2004;37:66-74.

[176] CADDEN A-M. Comparative Effects of Particle Size Reduction on Physical Structure and Water Binding Properties of Several Plant Fibers. Journal of food science. 1987;52:1595-9.

[177] Jacobs PJ, Hemdane S, Dornez E, Delcour JA, Courtin CM. Study of hydration properties of wheat bran as a function of particle size. Food chemistry. 2015;179:296-304.

[178] Niu M, Hou G, Lee B, Chen Z. Effects of fine grinding of millfeeds on the quality attributes of reconstituted whole-wheat flour and its raw noodle products. LWT - Food Science and Technology. 2014;57:58-64.

[179] F. Guillon MC. Structural and physical properties of dietary fibres, and consequences of processing on human physiology. Food Research International 2000;33:233-45.

[180] Pietrasik Z, Janz JA. Influence of freezing and thawing on the hydration characteristics, quality, and consumer acceptance of whole muscle beef injected with solutions of salt and phosphate. Meat science. 2009;81:523-32.

[181] Wang Z, Sun Y, Dang Y, Cao J, Pan D, Guo Y, et al. Water-insoluble dietary fibers from oats enhance gel properties of duck myofibrillar proteins. Food chemistry. 2021;344:128690.

[182] C.M. Crehan EH, D.J. Troy ,D.J. Buckley. Effects of fat level and maltodextrin on the functional properties of frankfurters formulated with 5, 12 and 30 fat. Meat science. 2000;55: 463–9.

[183] Shuilian Wang Tc, Chenjie Wang, Liu Shi, Wei Wang, Hong Yang， Min Cui. Effect of particle sizes of soy okara on textural,color, sensory and rheological properties of pork meat gels. Journal of Food Quality. 2015: 248–55.

[184] Huang SC, Shiau CY, Liu TE, Chu CL, Hwang DF. Effects of rice bran on sensory and physico-chemical properties of emulsified pork meatballs. Meat science. 2005;70:613-9.

[185] Sánchez-Alonso I, Solas MT, Borderías AJ. Technological implications of addition of wheat dietary fibre to giant squid (Dosidicus gigas) surimi gels. Journal of Food Engineering. 2007;81:404-11.

[186] Li C, Liu D, Zhou G, Xu X, Qi J, Shi P, et al. Meat quality and cooking attributes of thawed pork with different low field NMR T(21). Meat science. 2012;92:79-83.

[187] Li K, Liu J-Y, Bai Y-H, Zhao Y-Y, Zhang Y-Y, Li J-G, et al. Effect of bamboo shoot dietary fiber on gel quality, thermal stability and secondary structure changes of pork salt-soluble proteins. CyTA - Journal of Food. 2019;17:706-15.

[188] Nan H, Stepanova T, Li B, Kondratiuk N. Effect of Agaricus bisporus on gel properties and microstructure of chicken batters. Journal of Hygienic Engineering and Design. 2021;36:170-8.

[189] Nan H, Zhou H, Li B, Stepanova T, Kondratiuk N. Effects of Agaricus bisporus alone or in combination with soybean oil or water as fat substitutes on gel properties, rheology, water distribution, and microstructure of chicken batters. Food Science and Technology. 2022;42:e116121.

[190] Hu H, Pereira J, Xing L, Zhou G, Zhang W. Thermal gelation and microstructural properties of myofibrillar protein gel with the incorporation of regenerated cellulose. Lwt. 2017;86:14-9.

[191] Choi YS, Kim YB, Hwang KE, Song DH, Ham YK, Kim HW, et al. Effect of apple pomace fiber and pork fat levels on quality characteristics of uncured, reduced-fat chicken sausages. Poultry science. 2016;95:1465-71.

[192] Zhou Y, Dai H, Ma L, Yu Y, Zhu H, Wang H, et al. Effect and mechanism of psyllium husk (Plantago ovata) on myofibrillar protein gelation. Lwt. 2021;138:110651.

[193] Xiao H, Wu Y, Liu J, Zhou L, Zeng X, Zhao X. Potential mechanism of different gelation properties of white and red muscle fibre from crocodile (Crocodylus siamensis) meat: Study of myofibrillar protein. Lwt. 2021;143:111045.

[194] Pereira J, Sathuvan M, Lorenzo JM, Boateng EF, Brohi SA, Zhang W. Insight into the effects of coconut kernel fiber on the functional and microstructural properties of myofibrillar protein gel system. Lwt. 2021;138:110745.

[195] Cen S, Zhang L, Liu L, Lou Q, Wang C, Huang T. Phosphorylation modification on functional and structural properties of fish gelatin: The effects of phosphate contents. Food chemistry. 2022;380:132209.

[196] Ren Z, Chen Z, Zhang Y, Lin X, Li Z, Weng W, et al. Effect of heat-treated tea water-insoluble protein nanoparticles on the characteristics of Pickering emulsions. Lwt. 2021;149:111999.

[197] Ren Z, Cui Y, Wang Y, Shi L, Yang S, Hao G, et al. Effect of ionic strength on the structural properties and emulsion characteristics of myofibrillar proteins from hairtail (Trichiurus haumela). Food Research International. 2022;157:111248.

[198] Cofrades S, Benedí J, Garcimartin A, Sánchez-Muniz FJ, Jimenez-Colmenero F. A comprehensive approach to formulation of seaweed-enriched meat products: From technological development to assessment of healthy properties. Food Research International. 2017;99:1084-94.

[199] Marti-Quijal FJ, Zamuz S, Tomasevic I, Rocchetti G, Lucini L, Marszalek K, et al. A chemometric approach to evaluate the impact of pulses, Chlorella and Spirulina on proximate composition, amino acid, and physicochemical properties of turkey burgers. J Sci Food Agric. 2019;99:3672-80.

[200] Bach F, Helm CV, Bellettini MB, Maciel GM, Haminiuk CWI. Edible mushrooms: a potential source of essential amino acids, glucans and minerals. International Journal of Food Science & Technology. 2017;52:2382-92.

[201] Ceron-Guevara MI, Rangel-Vargas E, Lorenzo JM, Bermudez R, Pateiro M, Rodriguez JA, et al. Reduction of Salt and Fat in Frankfurter Sausages by Addition of Agaricus

bisporus and Pleurotus ostreatus Flour. Foods. 2020;9.

[202] Jurić S, Jurić M, Siddique MAB, Fathi M. Vegetable Oils Rich in Polyunsaturated Fatty Acids: Nanoencapsulation Methods and Stability Enhancement. Food Reviews International. 2020;38:32-69.

[203] Flávia Santi Stefanello, Mariana dos Santos da Silva, Carlos Pasqualin Cavalheiro, Leadir Lucy Martins Fries, Fernanda Luísa Ludtke, Ernesto Hashime Kubota, Carlos Pasqualin Cavalheiro, eadir Lucy Martins Fries, Fernanda Luísa Ludtke, Ernesto Hashime Kubota. Oxidative and microbiological stability of fresh pork sausage with added sun mushroom powder. Ciênc Agrotec, Lavras. 2015;39:381-9.

[204] Pirjo Mattila KSa, MSc,Vieno Piironen. Functional properties of edible mushrooms. Nutrition. 2000;16:694-6.

[205] Alnoumani H, Ataman ZA, Were L. Lipid and protein antioxidant capacity of dried Agaricus bisporus in salted cooked ground beef. Meat science. 2017;129:9-19.

[206] Papastergiadis A, Mubiru E, Van Langenhove H, De Meulenaer B. Malondialdehyde measurement in oxidized foods: evaluation of the spectrophotometric thiobarbituric acid reactive substances (TBARS) test in various foods. Journal of agricultural and food chemistry. 2012;60:9589-94.

[207] Cerón-Guevara MI, Santos EM, Lorenzo JM, Pateiro M, Bermúdez-Piedra R, Rodríguez JA, et al. Partial replacement of fat and salt in liver pâté by addition of Agaricus bisporus and Pleurotus ostreatus flour. International Journal of Food Science & Technology. 2021;2:1-11.

APPENDICES

APPENDIX A

OPERATION KEY POINTS

NO: 2021061601

Production process and operation key points of produce low-fat chicken sausage with *Agaricus bisporus* mushroom

Version number A

Date posted 20210616

Maker Haijuan Nan

Unit Henan Jiaduoduo Food Co., Ltd.

China Henan

Henan Jiaduoduo Food Co., Ltd.

APPENDIX B

<table>
<tr><td rowspan="2">HJF
Controlled document</td><td colspan="2">NO:2021061601</td></tr>
<tr><td colspan="2">Commencement date :20210616</td></tr>
<tr><td>Production process and operation key points of produce low-fat chicken sausage with Agaricus bisporus mushroom</td><td>Page 1</td><td>Total of 5 pages</td></tr>
</table>

1. Objective:

Standardize product production and ensure product quality.

2. Application scope:

It is suitable for the low-fat chicken sausage with *Agaricus bisporus* mushroom in meat products factory of Jiaduoduo Food Co., Ltd.

3. Product formula:

All the raw chicken batters are prepared with 100 kg chicken breast meat, 13.33 kg pork back-fat, 33.33 kg ice water, 6.67 kg *Agaricus bisporus* mushroom powder, 13.33 kg soybean oil, 2.33 kg NaCl, 0.50 kg Sodium tripolyphosphate, 0.17 kg chicken essence, 1.08 kg sugar, 0.25 kg ground white pepper, 8.33 kg potato starch. Therein, the raw chicken breast meat (Arbor Acres, females, 49-day-old; moisture $< 73\%$, protein $> 21\%$: pH, 5.65-5.90); *Agaricus bisporus* mushroom powder (moisture $\leq 7\%$. Sieved down through a 120 mesh sieve). Soybean oil (first grade).

APPENDIX C

HJF Controlled document	NO:2021061601	
	Commencement date :20210616	
Production process and operation key points of produce low-fat chicken sausage with *Agaricus bisporus* mushroom	Page 2	Total of 5 pages

4. Process content

4.1 Technological process:

Raw materials receive → Thaw → Dressing → Ground → Chopping* → Filling → Baking → Hot processing* → Packaging → Sterilization → Cooling → Labeling → Packing→Warehousing.

Note: * indicates the key process.

4.2 Raw material reception and dressing:

Requirements for the selection of fresh (frozen) chicken breasts and pork back fat as raw materials. Frozen chicken breasts and pork back fat should be well frozen, thawed at 18+2 °C, and the center temperature should be controlled at 0-4 °C after thawing. Fresh chicken breasts and pork back fat should be pre-cooled to an internal temperature of 4 °C before use. After thawing, chicken breasts and pork back fat should be repaired timely to remove blood stasis, floating hair, broken bones, impurities, etc, and the temperature of chicken breasts and pork back fat

APPENDIX D

HJF	NO:2021061601	
Controlled document	Commencement date :20210616	
Production process and operation key points of produce low-fat chicken sausage with *Agaricus bisporus* mushroom	Page 3	Total of 5 pages

should be controlled within 0-4 °C. *Agaricus bisporus* powder was mixed with soybean oil at a ratio of 1:2 for 24 hours to absorb oil and expand.

4.3 Ground

After removing fat and other non-musclar material, the chicken breasts and pork back fat should be ground using a meat grinder with a 6 mm holes plate (MGB-120, China) respectively. The meat grinder should be in proper operation with sharp blades, thus ensuring that the granularity of the chicken breasts and pork back fat is evident.

4.4 Chopping

The ground chicken breasts (0–4°C) should be chopped with salt, tripolyphosphate, and 1/3 ice water by a chopper at 1500 rpm for 60 seconds. After a pause of 3 minutes, ground chicken should be chopped with ground pork back fat, pre-mix of *Agaricus bisporus* powder and soybean oil, sugar, ground white pepper, chicken essence, potato starch, and 1/3 ice water at 1500 rpm for 120s. After a further pause of 3

APPENDIX E

<table>
<tr><td colspan="2">HJF
Controlled document</td><td colspan="2">NO:2021061601
Commencement date :20210616</td></tr>
<tr><td colspan="2">Production process and operation key points of produce low-fat chicken sausage with Agaricus bisporus mushroom</td><td>Page 4</td><td>Total of 5 pages</td></tr>
</table>

minutes, the ground chicken should be chopped with the remaining 1/3 ice water at 3000 rpm for 60 seconds. During the chopping process, it is necessary that the center temperature of the mixture is always lower than 10°C.

4.5 Filling

Immediately after chopping, the batter should be stuffed by a vacuum stuffer in 18 mm diameter edible collagen sausage casings. Chicken sausages should be linked automatically at 10 cm intervals.

4.6 Baking, hot processing

The uncooked sausages should be baked at 60 °C for 30 min and then in a water bath at 80 °C for 30 min until the internal temperature is 72 °C.

4.7 Vacuum packaging, sterilization, and cooling

Vacuum pack the cooked sausages and heat them in a water bath at

APPENDIX F

HJF Controlled document	NO:2021061601 Commencement date :20210616	
Production process and operation key points of produce low-fat chicken sausage with *Agaricus bisporus* mushroom	Page 5	Total of 5 pages

80°C for 15 seconds. Immediately cool the cooked sausages to 20 °C with tap water and store at 4 °C.

4. 8 Labeling, packing and warehousing

After the surface of the product packaging has dried, it should be correctly and securely labelled and the net content of the whole box must not be negatively deviated. Each cardboard box contains 50 sausages. The packaged products should be stored in a warehouse at 0 - 4°C and have a shelf life of 30 days.

APPENDIX G

CERTIFICATE OF PRODUCT APPLICATION

Henan Jiaduoduo Food Co., Ltd. conducted pilot tests on the low-fat chicken sausage products containing *Agaricus bisporus* mushrooms with consumer characteristics developed by Nan Haijuan from 2021 to 2022. Furthermore, the company put the product on the market in 2022 for sale, which is deeply loved by consumers and had generated over $300,000 in direct economic benefits for the company.

Henan Jiaduoduo Food Co., Ltd.

December 2022

APPENDIX H

CERTIFICATE OF PRODUCT APPLICATION

Henan Fengxiang Fengwei Food Co., LTD conducted pilot tests on the low-fat chicken sausage products containing *Agaricus bisporus* mushrooms with consumer characteristics developed by Nan Haijuan from 2021 to 2022. Furthermore, the company launched the products into the market in 2022, which is popular among consumers, and gained a direct economic benefit of $20,000.

Henan Fengxiang Fengwei Food Co., LTD

December 2022