IntechOpen

Intensive Animal Farming

A Cost-Effective Tactic

Edited by Shumaila Manzoor and Muhammad Abubakar

Intensive Animal Farming - A Cost-Effective Tactic

Edited by Shumaila Manzoor and Muhammad Abubakar

Published in London, United Kingdom

Intensive Animal Farming - A Cost-Effective Tactic
http://dx.doi.org/10.5772/intechopen.100887
Edited by Shumaila Manzoor and Muhammad Abubakar

Contributors
Muhammad Bilal, Ken Tasaki, Abigail N. Tasaki, Grace Opadoyin Tona, Sami Mili, Rym Ennouri, Manel Fatnassi, Hajer Zarrouk, Rabeb Thabet, Houcine Laouar, Rafael Alexandre Muchanga, Lopes C. César Mugabe, Maicon Pereira Lents, Emmanuel Emydio Gomes Pinheiro, Muhammed Nurye Gebeyehu, Douglas Landblom, Songul Senturklu, Gerald Stokka, Larry Cihacek, Shumaila Manzoor, Muhammad Abubakar, Zainab Syed

First published in London, United Kingdom, 2023 by IntechOpen
IntechOpen is the global imprint of INTECHOPEN LIMITED, registered in England and Wales, registration number: 11086078, 5 Princes Gate Court, London, SW7 2QJ, United Kingdom

British Library Cataloguing-in-Publication Data
A catalogue record for this book is available from the British Library

Additional hard and PDF copies can be obtained from orders@intechopen.com

Intensive Animal Farming - A Cost-Effective Tactic
Edited by Shumaila Manzoor and Muhammad Abubakar
p. cm.
Print ISBN 978-1-80356-101-1
Online ISBN 978-1-80356-102-8
eBook (PDF) ISBN 978-1-80356-103-5

Meet the editors

Dr. Shumaila Manzoor, a scientist and laboratory worker from the National Veterinary Laboratory, Islamabad, Pakistan, has more than 12 years of experience in various areas of veterinary disease diagnosis. Her expertise on transboundary animal diseases (TADs) is well recognized, and she has worked with the Food and Agriculture Organization (FAO) control programs of TADs in the country. She has conducted various trainings for field as well as laboratory staff. She has published research papers, review articles, and book chapters on different areas of veterinary disease diagnosis and control. She has been a peer reviewer for the *Research Journal for Veterinary Practitioners and Veterinary Sciences: Research and Reviews* for more than five years.

Dr. Muhammad Abubakar, a scientist from the National Veterinary Laboratory, Islamabad, Pakistan, has more than 17 years of experience in various areas of veterinary sciences. His expertise on transboundary animal diseases (TADs) at national and international levels is well known. He has established various diagnostic laboratories for the diagnosis of TADs. He has been involved in organizing and conducting training for field and laboratory staff. He has published numerous research papers, review articles, and book chapters in veterinary sciences. He has also worked in academia, teaching and supervising research at graduate and undergraduate levels. He is currently editor in-chief of the *Research Journal for Veterinary Practitioners* and *Veterinary Sciences: Research and Reviews*.

Contents

Preface

Intensive animal farming, also known as factory farming, is an advanced technique to maximize production while reducing the cost of production. Intensive animal farming is a very recent development in agribusiness, which involves the high stocking of animals like cattle, poultry, and fish on large scales and the use of advanced biotechnology to enhance production.

This book consists of three main sections.

The first section is about livestock and poultry farming. Livestock, particularly cattle and poultry, are important contributors to total food production in the world. Intensive animal farming is very important from a food security perspective.

The second section discusses fish farming and different aspects of aquaculture such as rearing systems, nutrition, and diseases at the farm level.

The third section contains information regarding innovations and advances in technology. Intensive animal farming is very prevalent in developed countries and aims to produce large quantities of milk, meat, and eggs at low cost. Intensive farming involves mass production through modernized feeding systems and improved breeding and health programs.

I want to express my gratitude and thanks to all the contributing authors for their hard work. I want to thank Sara Tikel, Lucija Tomicic, and other staff at IntechOpen for their efforts to bring about the final product. I want to acknowledge my colleagues, friends, and family, especially my son, for all their support.

Shumaila Manzoor and Muhammad Abubakar
National Veterinary Laboratory,
Islamabad, Pakistan

Section 1

Livestock and Poultry Farming

Chapter 1

Global Perspectives of Intensive Animal Farming and Its Applications

Shumaila Manzoor, Zainab Syed and Muhammad Abubabakar

Abstract

Agricultural farming outputs are dependent upon the production type because different farming systems create different products. Intensive animal farming is widely used for the production of products that have societal importance, including meat, milk, wool, leather, fur, eggs, and honey. To ensure their timely production with limited cost, advanced technological processes, and chemicals (pesticides, herbicides, and fertilizers in large amount) are carried out in this intensive factory farming. Amongst animal farming, the livestock sector is the fastest-growing agricultural sector. The global shift toward intensive animal farming for high productivity yield has rendered a negative impact on the environment and biodiversity and is now an alarming sign for global warming. It has also resulted in soil, water, and air pollution due to the emission of greenhouse gases from the waste generated by these animals. Rapid use of antimicrobials in these farming systems has led to the emergence of drug-resistant pathogens. Therefore, an integrated and comprehensive approach covering the nonmarket outputs of the farming system is required for monitoring these global trends.

Keywords: intensive animal farming, factory farming, high production, technological processes, global warming

1. Introduction

Intensive animal farming [1, 2] or livestock farming is an intensive agriculture type that is destined to increase animal production by providing them all necessities and a favorable environment while reducing the rearing cost [3]. The environment provided to the animals here includes nutrition, shelter, water, optimum temperature and humidity, and veterinary management. It is also known as factory farming [4].

The term Factory farming means "any globally adopted farming system where flock of animals are kept under one roof in a confined setting, that is, a cage or stall" [5]. According to the United States Department of Agriculture (USDA), "a farm with 1000 cattle or 125,000 chickens is referred as factory farming system" [6]. However, according to the European Union (EU), "farm carrying 40,000 chickens is referred to as factory farm or macro-farm" [7]. The products of the animal farming industry are milk, meat, egg, and other animal products which are readily available for human consumption and are much-liked food amongst people across the globe. Feeding the entire world is the sustainable development challenge in the coming few years. Meat plays a major role in this. The demand for meat has increased rapidly over the past 50 years and it has tripled now [8]. According to the estimates of Food and Agriculture Organization of the United Nations (FAO), approximately 68.1 billion animals were slaughtered in 2012 for meat [9]. However, this figure increased to 80 billion in 2018. Poultry meat is the most popular meat worldwide [8].

Based on the above estimates, every individual on Earth is provided with an average of 42.9 kg of meat. In developed nations, average 76.2 kg of meat is consumed by individuals and, on the contrary, in developing nations, 33.4 kg of meat is consumed by individual on average basis [10]. Asia (42.1%) is the largest producer of livestock followed by America (31.4%), Europe (19.0%) [11], and Africa (5.5%). Besides meat, animals also provided us with milk (5.7 billion tons) and eggs (72 million tons) [11]. Chicken laid 1.25 billion eggs; other poultry laid about 87 million eggs. Much of the animals' products discussed above came from those animals who were raised by someone else on our behalf and amongst them, the majority were reared using intensive animal farming [11]. Factory, intensive, industrial animal farming, and concentrated animal feeding operation (CAFO) all are used for a modern form of intensively rearing of fowl, fish for their various edible products, including their meat (i.e., flesh and fat) and other forms of animal protein (i.e., dairy and eggs). Intensive farming can also be defined as an economic pursuit involving domestic animals for human uses such as obtaining honey, fur, leather, wool, and fertilizer. The sole purpose of this type of farming is to ensure maximum production with maximum profit [12]. According to Archambeaud [13], intensive farming is a farming type where agricultural machinery is employed for achieving higher productivity, that is, the excessive usage of pesticides, fertilizer, or disease or weed-resistant chemicals. This high productivity yield also renders a negative impact on the environment and biodiversity [13] which will be discussed later in this chapter.

2. History

Intensive animal farming is the most recent advancement in agriculture history which is also the result of scientific and technological developments. In the late nineteenth century, innovations were made in mass production. Later, in early twentieth century; vitamin discovery and their associated role in animal nutrition was the hallmark of Industrial Revolution because it allowed poultry to be raised at the domestic level [14]. Moving further, antibiotics and vaccines discovery have further lightened the livestock by reducing the number of disease-causing pathogens [15]. Chemicals used in World War II gave the idea of synthetic pesticide discovery [14]. The development of transport networks and technology enabled the distribution of agricultural products over long distances.

The era of high-put farming began in Britain in 1947, when the new Agriculture Bill provided subsidies to farmers to promote more production by introducing new technologies to reduce Britain's dependence on imported meat. According to United Nations "the intensification of livestock production" was found to ensure food security [16]. In 1966, the United States, Great Britain, and other developed countries began large-scale farming of beef and dairy cattle and domestic pigs [17]. From the heartland of America and Western Europe, factory farming became globalized in the later twentieth century and continues to expand, replacing traditional livestock farming practices in an increasing number of countries. In 1990, intensive animal husbandry accounted for 30% of world meat production, and by 2005 this had risen to 40% [17]. Worldwide meat production in 2020 was 328 million tons which suggests that the demand for meat has reached 90% [18].

3. Global perspectives

Globally, more than 70 billion animals are slaughtered every year for food. On the basis of data provided by UN FAO, the five major groups of animals slaughtered are cows, chicken, goats, sheep, and pigs [19]. It is expected that by 2050, intensive farming production will double with the major advancement taking place in less industrialized countries. This expansion has had serious consequences because only the livestock sector generates about 18% of greenhouse gas which is more than any kind of transport. Moreover, 70% of the Earth's surface is directly or indirectly involved in livestock production leading to land degradation, environmental pollution, and other health-associated issues [20]. These problems will not go away on their own if more and more extensive farming systems are being shifted toward intensive animal farms where animals are raised in confinement at high stocking density using advanced machinery and biotechnology. Intensive farming production systems were the norms of Europe and America, but now this practice is increasingly becoming common in Asia and Latin America. According to UN reports, the global shift of farming systems and environmental problems associated with these systems has not given much importance which is why they now have become a serious concern [21].

There are numerous problems associated with intensive animal farming system, a few of which is discussed below:

3.1 Increased emission

As in intensive farming system, animals are raised indoors, therefore large amount of energy is required for heating, cooling, and ventilation as well as for feed production and transportation producing more emissions of carbon dioxide and anthropogenic nitrous oxide (which stays in the atmosphere for up to 15 years) and has more global warming potential leading to depletion of ozone layer. The livestock sector generates 64% ammonia emissions thereby contributing to acid rain and acidification of biodiversity [22].

Worldwide, farm animals are also a source of methane emission. Methane has 23 times more global warming potential than carbon dioxide. The operation of intensive farm animals leads to increased emission of methane because of animal diet which also causes diseases in animals and emits 50% more methane than animals feed on grasses in open lands. The only reason is that in intensive system animals are raised on a concentrated high-protein diet (made up of 50% corn and 80% soybean). This food is cheap and easy to produce and animals by eating such diets put on weight faster. This emission of greenhouse gases will continue to increase as the intensive farming system spread to more and more developing countries [22].

3.2 Climate changes

Developing countries suffer more from the impact of climate change because of abrupt increase in hunger and disease. This is because developing nations have limited coping capacities and they are dependent upon climate-sensitive food and water supply chains. Climatic changes are responsible for food scarcity in developing sectors. Excessive flooding, storms, loss in biodiversity, land degradation, and water and air pollution affect developing countries mostly because of health, poverty, and infrastructure constraints [23].

3.3 Loss in biodiversity

Animal waste and droppings are not treated properly, as farmers often dispose of their waste in rivers, where they pollute the water and impend the biodiversity of the ecosystem. Waste products of poultry emit ammonia and nitrous oxide leading to nitrogen pollution of water and soil [24]. Uneven use of pesticides and fertilizers can also pollute soil, water, and air [25]. The continuous degradation of environment and loss in biodiversity is an alarming sign for global warming [25].

3.4 Land degradation

Land degradation due to deforestation is also one of the major problems. Animals reared for meat, milk, and eggs production are already covering one-third of the Earth planet surface. Cattle ranching is the primary cause of deforestation because forested lands are cut and cleared for making proper room for animal grazing and meat production [26]. The meat thus produced is exported to developed countries. The high meat demand in developed countries is rendering negative impact on the meat-producing country both due to deforestation and soil erosion. Besides grazing, high-quality protein feed production is also putting pressure on land. The number of those protein diets continues to increase as intensive farming increases. The employment of large area of land for feed production is resulting in loss of biodiversity, soil erosion, and increased greenhouse gases emission [27].

3.5 Antimicrobial resistance

Increasing demand for animal protein in intensive farming system has led to an increase in antimicrobial use (AMU) leading to the emergence and spread of antimicrobial resistance (AMR) [28]. Antimicrobials are mostly used in intensive animal farming to prevent or treat infection and are also given in animal diets for rapid growth [29]. Previous reported literature has shown that 73% of the antimicrobials available locally are given to animals raised for food. AMU in food-producing animals can also affect humans, leading to antimicrobial resistance. The widespread use of antimicrobials in farms can also contaminate the environment, leading to the emergence of drug-resistant pathogens. Therefore, monitoring the global trends of antimicrobial use in intensive farming system is important to track progress associated with antimicrobial stewardship programs carried across regions [28].

4. Types of livestock farming

Based on the production processes, livestock farming is of different types which are described below in detail:

4.1 Intensive animal farming

It is also known as conventional or high-put farming system [2]. In intensive animal farming, to ensure healthy and faster animal production, animals are housed with adequate nutrients, feed, and temperature. Breed selection in this system is made up of different production types. It is both labor and capital-intensive. The primary goal of intensive farming is the attainment of high production [30].

4.2 Semi-intensive animal farming

In semi-intensive farming mode, animals are housed and fed but they are allowed to move or graze around the farm to forage within a confined area inside the farm premises [31].

4.3 Extensive animal farming

It is also known as a low-intensity or low-input farming system. In extensive farming systems, rearing is carried out on open areas of the lands, that is, pastures, meadows, and mountains so that animals can get maximum benefit from the natural products. The farming system is applicable to the animals who are intended to be adapted to the field [32]. This system supports the preservation of the ecological unit. In this type of farming, external resources like pesticides and fertilizers are used in low quantity [33].

4.4 Organic animal farming

Organic farming is a type of animal farming system whose primary aim is to produce high-quality food without the use of synthetic chemicals, that is, chemical fertilizers or pesticides, etc. Additionally, animals are grown in open spaces and fed on natural resources [34].

Livestock represents all animal types like buffaloes, cattle, goats, sheep, horses, and pigs, etc. and they are reared primarily for milk, meat, and wool production. Livestock farming is associated with the production of eggs, milk, and meat from domesticated animals [35].

The basic purpose of these farming systems is the production of agricultural products such as cereal, crops, rice, sheep, fish, and fowl. These all-farming systems are dependent on plants as their primary food source, which in turn, rely on the soil. Merely, the production of farming is influenced by the type of farming system and agricultural action chosen. **Figure 1** [36] depicts the typological classification of the farming system.

5. Pros and cons of intensive animal farming

Cattle farming has been an important part of society for years, ever seeing that people started domesticating animals to improve the quality of their life. However, as with most forms of farming, inclusive of agriculture, cattle farming too has strengthened, specifically in current many years. This has made livestock products more easily available and cheaper to buy; that is especially important in case one assumes that staples along with milk, honey, eggs, and meat are all merchandise in cattle farming [37].

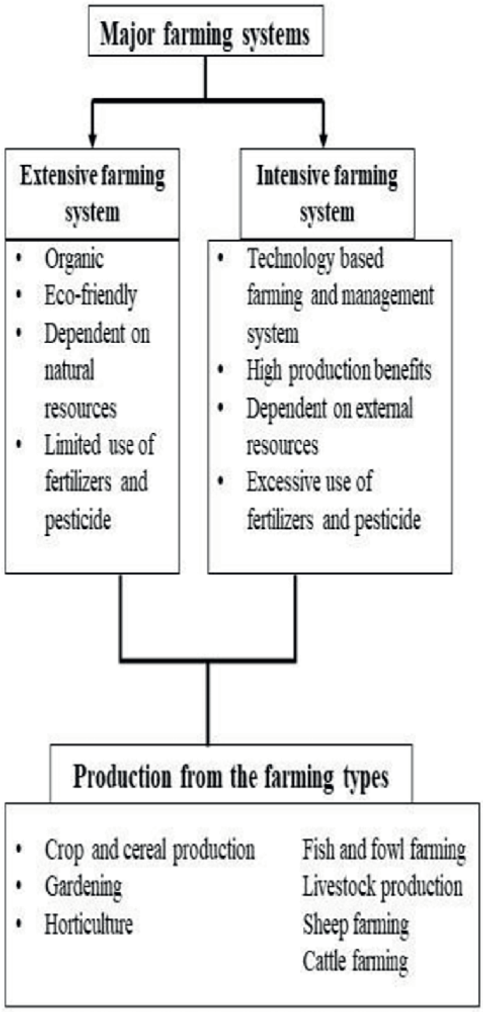

Figure 1.
Typological classification of major farming systems [36].

However, intensive livestock farming practices have on several occasions raised major concerns regarding food protection, animal welfare, and environmental effects—to the extent that cattle farming is often called "factory farming" [38].

5.1 Pros of intensive animal farming

The contribution of livestock farming to the country GPD (Gross domestic product) is about 883 billion dollars, but this amount does not include the services availed from retailers, butchers, and transport units and supplied to feed producers and equipment producers. Besides its role in economic development, the livestock sector increases the life expectancy of individuals by providing food security to about 1.3 billion people. Nowadays, it is the fastest-growing agricultural sector of the country's economy [39].

Intensive animal farming	
Advantages	**Disadvantages**
Cheaper and better-quality food products	Cost of external resources (pesticides and fertilizers)
Rational use of land	Cost of technical equipment
Limited manual work	Trained personnel are needed for technological operations
Faster production using the modern technology	Machinery replaces labor; hence less people are involved in agricultural operations
Global food safety and security	Damage landscape, environment, soil, and wild-life biodiversity

Table 1.
Advantages and disadvantages of intensive animal farming [40].

Intensive animal farming has been made possible by farming management practices that have helped to increase yield and production while bringing down the cost at a confined place. For example, farming units employing the practice of concentrated animal feeding operation (CAFO) have enabled farm owners to rear more animals in a restricted area, thereby maximizing the land potential [39].

5.2 Cons of intensive animal farming

Though animal farming has efficiently increased the production of animal products at a limited cost, yet the external products (cost-saving techniques) that have been used for ensuring the steady production of products have negatively impacted health and the environment [39].

In a confined environment, where flocks of animals are kept under one roof has increased the chances of animals becoming more susceptible to diseases. In low- and middle-income countries (LMIC), livestock diseases have been transmitted to about 2.4 billion humans. To reduce the burden of zoonotic diseases, farmers frequently administered antibiotics to their animals leading to the evolution of drug-resistant pathogens [39]. Besides this, some farmers kept animals to live in stressful conditions. Unfortunately, practices persist where animals are transported long distances to the market in inhumane conditions or slaughtered in painful ways.

Keeping in view the above-described pros and cons of intensive animal farming, a few more advantages and disadvantages are listed in the **Table 1** given below:

6. Methodology

6.1 Guidelines

A meta-analysis-based study was designed to review the intensive animal farming system in Pakistan. The study was carried out according to the guidelines of "Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRIMA)" (Page et al., 2021).

Source	Number of Articles
Google Scholar	300
Google	10
Total	310

Table 2.
Number of articles obtained from searching international databases.

6.2 Literature search

An online search of international database sources, that is, Google Scholar, Research Gate, and Google was carried out to identify relevant studies reported from Pakistan from 2015 to 2023. A total of 310 searches were carried out. The reference list of the searched studies was further reviewed for any relevant publication. The duplicate article found using the above-mentioned databases was removed using EndnoteX7 (Thomson Reuters, New York, NY, USA). The searched sources of 500 articles are given in **Table 2**.

6.3 Keywords

The following keywords were searched: Livestock farming, intensive animal farming, and application of intensive animal farming in Pakistan.

6.4 Studies inclusion criteria

The eligible studies were selected for inclusion in this meta-analysis when the following criteria were met. (1). Full-text articles available in English language. (2). Studies reported from different regions of Pakistan. (3). Cross-sectional and retrospective studies. (4). Sample size provided. (5). Studies reporting the effect of climatic changes and antimicrobial use in livestock.

6.5 Studies exclusion criteria

Studies were excluded based on the following exclusion criteria: (1). Duplicated data or review articles and conference abstracts. (2). Articles without full text. (3). Articles with abstract only. (4). Data reported from other regions of the world. (5). Research articles conducted before 2015.

6.6 Comparison of intensive and extensive farming system

For better understanding of the advantages of intensive farming system, a comparison was undertaken for analyzing the efficiency of intensive and extensive farming system. Different farming practices, that is, amount of milk production and farming technical efficiency were measured between the two farming practices.

6.7 Statistical analysis

This meta-analysis was computed using random effects model with Open-Meta Analyst version 10.10. The heterogeneity of the studies was checked using Cochran's

Q test. The variation across studies was observed by the forest plot as well as the inverse variance index (I2). Values of I2 (25, 50, and 75%) were considered as low, medium, or high heterogeneity, respectively. In this meta-analysis, the heterogeneity value was >75%, therefore, the DerSimonian and Laird random effects models with 95% CI. Funnel plot analysis was carried out if heterogeneity was of moderate to high level. Subgroup meta-analyses were then employed by publication year.

7. Results

7.1 Selection of studies

The aim of the present study was to determine intensive animal farming practices employed in Pakistan and investigate the effect of different factors on livestock production. Different international databases, including Google Scholar and Google, were searched (from 2015 to 2023) to identify studies that addressed the intensive animal farming activities in Pakistan.

For this meta-analysis, a total of 310 articles were identified in the initial search. Out of 310 searches, 254 articles were excluded because of their irrelevance and not being reported from Pakistan. Case reports, conference abstracts, and review articles were also excluded. 50 articles were also excluded for reasons of being duplicates, titles and not having full text. Six relevant articles were selected because they met the inclusion criteria, and their full texts were reviewed. The flow diagram of the selection process of the included studies is shown in **Figure 2**.

The characteristic of the included study is shown in **Table 3** given below:

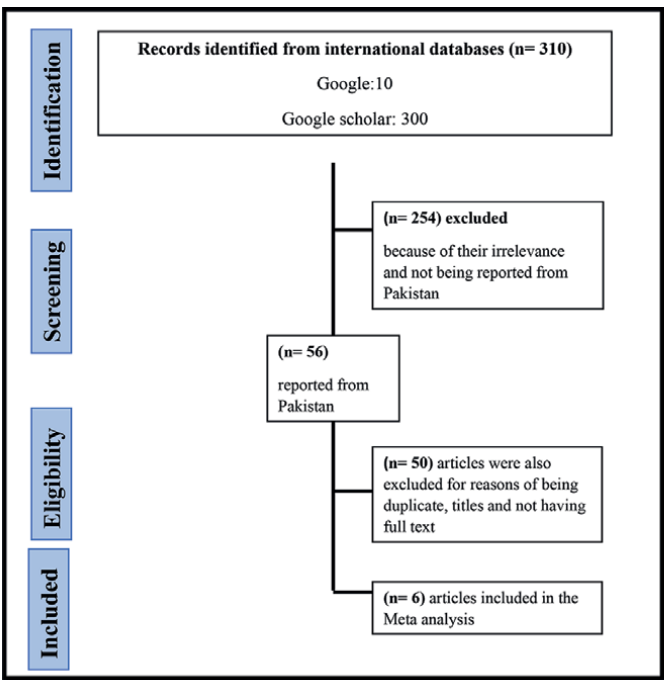

Figure 2.
Flow diagram showing selection criteria of the selected studies.

Researcher	Year	Study aim	Country	Results	Conclusion	Study Weight	Reference
Umair et al.,	2020	To determine trends of antimicrobial use in dairy farm.	Punjab, Pakistan	Defined daily dose was 47.71 out of 1000 cows tested.	Increased antimicrobial usage in dairy sector.	16.87%	[41]
Abid et al.,	2016	Impact of climate change on 450 farms.	Punjab, Pakistan	55% were vulnerable to extreme temperature, and insect attack, and 35% are vulnerable to soil problems.	Limited resources and lack of infrastructure are contributing toward climate changes.	16.82%	[42]
Mohsin et al.,	2019	Use of medically important antimicrobial in food-producing animals.	Punjab, Pakistan	High consumption of antimicrobials was seen in 30 flocks. The annual use of medically important antimicrobials was 250.84 mg/kg.	The frequent antibiotics used were colistin, tylosin, doxycycline, and enrofloxacin.	16.13%	[43]
Habib et al.,	2016	Analysis of food supply in livestock.	Pakistan	Crop residues (58.8%) were the predominant food source for livestock followed by crude protein (37.2%).	Indigenous food sources were rarely available for livestock.	16.65%	[44]
Shahzad & Abdulai	2020	Impact of extreme climatic conditions on crop.	Punjab, Pakistan	540 farmers were interviewed. It was found that climate-related risks had (Extreme temperature and rainfall) severely impacted soil, crop rotation, and farmers' income.	Mitigation strategies should be adopted to cope with the situation.	16.84%	[45]
M. Riaz	2022	Livestock-integrated farming practices.	Faisalabad, Pakistan	105 responses were collected from farm owners. Livestock sector is dynamic for Pakistan's agriculture, contributing 17% to energy and 33% to protein consumption.	Farmers in integrated system were using traditional methods for milk production. Modern practices and procedures were not common in integrated rural systems.	16.67%	[46]

Table 3.
Traits of the studies included in the meta-analysis.

7.2 Forest plot

Significant heterogeneity values were observed in the forest plot built for intensive animal farming activities carried out in Pakistan. included studies The heterogeneity values of the included studies was (Tau^2 = 0.178, P = <0.001, I^2 of 99.17%), as shown in **Figure 3**.

For analyzing trends in intensive animal farming practices being overtaken in Pakistan, a subgroup analysis was performed based on year. As depicted in **Figure 4**, substantial heterogeneity was seen during the study period.

7.3 Comparison between intensive and extensive animal farming

The animal feeding pattern and milk production system were compared in a study carried out in Sri Lanka where the author concluded that animals reared in the intensive farming system had highest herd size (3.7 animal unit) with better feeding level when compared with extensive farming system (2.7 animal unit). The average milk production under extensive systems was significantly lower (P < 0.01) at 3.9 animal units per day compared to 5.41 animal units per day under the intensive farming system. Livestock farming is an important source of income for peoples of Sri

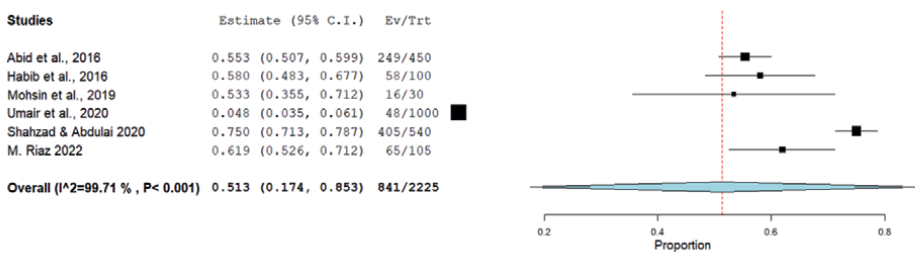

Figure 3.
Forest plot of intensive animal farming and its 95% confidence interval (CI). The pooled prevalence was calculated using a random-effect model. Ev/Trt = No. of VRSA positive isolates/Total no. of samples.

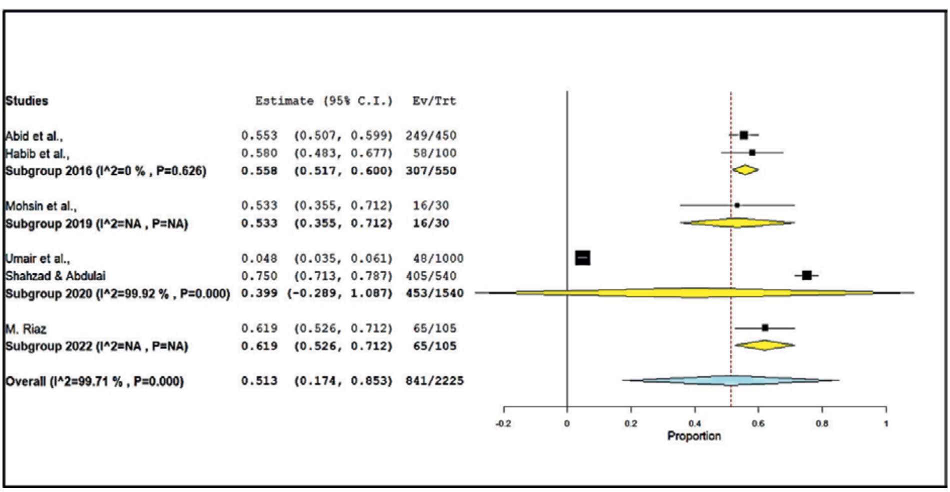

Figure 4.
Subgroup analysis of intensive animal farming carried out in Pakistan in different time periods.

Lanka and dairying under an intensive farming system is proven more profitable than an extensive farming system [47].

In Bangladesh, performance traits of buffalo selected randomly from 14 farms under both intensive and extensive systems were evaluated and it was found that dry milk yield and live weight were considerably higher in the intensive farming system. Reproductive traits were moderately higher under the intensive farming system. Intensive farming system is currently under application in Bangladesh for better milk production [48].

8. Conclusions

Intensive animal farming has both positive and negative impact on the environment and biodiversity depending upon the agricultural production. Intensive animal farming has provided society with marketable goods which can also be exported to foreign countries. The farming output uses market values which are limited because positive and negative outwardness are created along with the product goods and services in the agricultural activities. Therefore, the integrated/comprehensive approach covering market and nonmarket farming system output is required.

Acknowledgements

Replace the entirety of this text with acknowledgments. Usually, the acknowledgments section includes the names of people or institutions who in some way contributed to the work, but do not fit the criteria to be listed as the authors.

Conflict of interest

The authors declare no conflict of interest.

Author details

Shumaila Manzoor[1]*, Zainab Syed[2] and Muhammad Abubabakar[1]

1 National Veterinary Laboratory, Islamabad, Pakistan

2 Alama Iqbal Open University, Islamabad, Pakistan

*Address all correspondence to: smnvl786@gmail.com

References

[1] Lusk J. Why Industrial Farms Are Good for the Environment. Stillwater, Oklahoma, United States: NY Times; 2016

[2] Zhukova Y et al. Milk urea content and δ13C as potential tool for differentiation of milk from organic and conventional low-and high-input farming systems. Turkish Journal of Agriculture-Food Science and Technology. 2017;**5**(9):1044-1050

[3] White M. From Being to Thing: Personhood, Animalhood, and Deanimalization in the Human-Animal Relationship [Thesis]. Williamsburg, Virginia: William & Mary; 2018

[4] Mpofu I. Ecosystem services from different livestock management systems. In: The Role of Ecosystem Services in Sustainable Food Systems. Cambridge, United States: Elsevier; 2020. pp. 135-140

[5] Kondombo SR. Improvement of Village Chicken Production in a Mixed (Chicken-Ram) Farming System in Burkina Faso. Netherland: Wageningen University and Research; 2005

[6] Cockshaw R. The end of factory farming: Alternatives to improve sustainability, safety, and health. Voices in Bioethics. 2021;**7**:1

[7] Priekulis J, Melece L, Laurs A. Most appropriate measures for reducing ammonia emissions in Latvia's pig and poultry housing. Agronomy Research. 2019;**17**(3):797-805

[8] Ritchie H, Rosado P, Roser M. Meat and Dairy Production. Wales, England: Our World in Data; 2017

[9] FAO. FAO Statistical Yearbook 2012. Rome, Italy: FAO; 2012

[10] Tosun D, Demirbaş N. What are the problems of the red meat processing industry?: A case study from Turkey. Food Science and Technology. 2020;**41**:522-528

[11] Furber M. Wrongs & Responsibilities. Abu Dhabi, U.A.E: Tabah Foundation; 2017

[12] Simon DR. Meatonomics: How the Rigged Economics of Meat and Dairy Make you Consume Too Much–And How to Eat Better, Live longer, and Spend smarter. Massachusetts: Conari Press; 2013

[13] Archambeaud M. Conservation Agriculture, a Way to High Efficiency Farming Systems–A2C le Site de l'agriculture de Conservation. France: Agriculture Conservation; 2022

[14] Mitra S, Bhattacharya A, Roy S. The History of Livestock Farming and Future Perspective. United States: Biotechnological Advances for Precision Feeding; 2019

[15] Laxminarayan R et al. Antibiotic resistance—The need for global solutions. The Lancet Infectious Diseases. 2013;**13**(12):1057-1098

[16] Pluhar EB. Meat and morality: Alternatives to factory farming. Journal of Agricultural and Environmental Ethics. 2010;**23**(5):455-468

[17] Nierenberg D, Mastny L. Happier Meals: Rethinking the Global Meat Industry. Vol. 171. Washington D.C: Worldwatch Institute; 2005

[18] Coetzee K. Predicting a straight-line future. Farmer's Weekly. 2021;**2021**(21029):28-28

[19] Sanders B. Global Animal Slaughter Statistics and Charts. Olympia, Washington, United States: Faunalytics; 2018

[20] Ilea RC. Intensive livestock farming: Global trends, increased environmental concerns, and ethical solutions. Journal of Agricultural and Environmental Ethics. 2009;**22**:153-167

[21] Pelletier N, Tyedmers P. Forecasting potential global environmental costs of livestock production 2000-2050. Proceedings of the National Academy of Sciences. 2010;**107**(43):18371-18374

[22] Sakadevan K, Nguyen M-L. Livestock production and its impact on nutrient pollution and greenhouse gas emissions. Advances in Agronomy. 2017;**141**:147-184

[23] Rust J, Rust T. Climate change and livestock production: A review with emphasis on Africa. South African Journal of Animal Science. 2013;**43**(3):255-267

[24] Olivier S. CAFOs as Hotspots: Effect on Ecosystem Services and Needed Change in Environmental Leadership. North Carolina: DukeSpace; 2012

[25] Machovina B, Feeley KJ, Ripple WJ. Biodiversity conservation: The key is reducing meat consumption. Science of the Total Environment. 2015;**536**:419-431

[26] Temesgen G, Amare B, Silassie HG. Land degradation in Ethiopia: Causes, impacts and rehabilitation techniques. Journal of Environment and Earth Science. 2014;**4**(9):98-104

[27] Gashu K, Muchie Y. Rethink the interlink between land degradation and livelihood of rural communities in Chilga district, Northwest Ethiopia. Journal of Ecology and Environment. 2018;**42**:1-11

[28] Tiseo K et al. Global trends in antimicrobial use in food animals from 2017 to 2030. Antibiotics. 2020;**9**(12):918

[29] Lhermie G, Tauer LW, Gröhn YT. The farm cost of decreasing antimicrobial use in dairy production. PLoS One. 2018;**13**(3):e0194832

[30] Magne M-A et al. Managing animal diversity in livestock farming systems: Types, methods and benefits. INRA Productions Animales. 2019;**32**(2):263e-280e

[31] Salas MÁS et al. Assessing dairy goat welfare in intensive or semi-intensive farming conditions in Mexico. Journal of Dairy Science. 2021;**104**(5):6175-6184

[32] Poux X. Low input farming systems in Europe: What is at stake. In: Proceedings of the JRC Summer University Ranco. Low Input Farming Systems: An Opportunity to Develop Sustainable Agriculture. Luxembourg: Office for Official Publications of the European Communities; 2008. pp. 1-11

[33] Fess TL, Kotcon JB, Benedito VA. Crop breeding for low input agriculture: A sustainable response to feed a growing world population. Sustainability. 2011;**3**(10):1742-1772

[34] Sener S. Aspects of organic farming. In: Introduction and Application of Organic Fertilizers as Protectors of our Environment. New Castle, United Kingdom: Cambridge Scholars Publishing; 2022. p. 37

[35] Tripathi A et al. Production and consumption pattern of livestock products in Meghalaya and its implications in development of market strategies for the state producers. Indian Journal of Animal Sciences. 2019;**89**(1):113-116

[36] Novikova A, Startiene G. The advantages and disadvantages of intensive

and extensive farming activities. Research for Rural Development. 2018;**2**:139

[37] Blount WP. Intensive Livestock Farming. Amsterdam, Netherland: Elsevier; 2013

[38] Anjum FM, Arshad MS, Hussain S. Factory farming and halal ethics. In: The Halal Food Handbook. New York, United States: John Wiley & Sons; 2020. pp. 121-147

[39] Pillai M. Advantages and Disadvantages of Intensive Farming. Noida, India: Conserve Energy Future; 2008

[40] Novikova A, Startiene G. Analysis of farming system outputs and methods of their evaluation. Research for Rural Development. 2018;**2**:7

[41] Umair M et al. First case report on quantification of antimicrobial use in corporate dairy farms in Pakistan. Frontiers in Veterinary Science. 2020;**7**:575848

[42] Abid M et al. Climate change vulnerability, adaptation and risk perceptions at farm level in Punjab, Pakistan. Science of the Total Environment. 2016;**547**:447-460

[43] Mohsin M et al. Excessive use of medically important antimicrobials in food animals in Pakistan: A five-year surveillance survey. Global Health Action. 2019;**12**(Suppl 1):1697541

[44] Habib G et al. Assessment of feed supply and demand for livestock in Pakistan. Journal of Agricultural Science and Technology, A. 2016;**2016**(6):191-202

[45] Shahzad MF, Abdulai A. Adaptation to extreme weather conditions and farm performance in rural Pakistan. Agricultural Systems. 2020;**180**:102772

[46] Riaz M. Livestock integrated farming in rural area of Pakistan. In: International Conference on Improving Tropical Animal Production for Food Security (ITAPS 2021). Amsterdam, Netherland: Atlantis Press; 2022

[47] Premaratne S et al. Feeding Patterns and Milk Production of Small-Scale Dairy Farmers under Semi-Intensive and Extensive Cattle Management Systems in Sri Lanka. United Kingdom: UK Knowledge; 2019

[48] Momin M, Khan M, Miazi O. Performance traits of buffalo under extensive and semi-intensive Bathan system. Iranian Journal of Applied Animal Science. 2016;**6**(4):823-831

Chapter 2

Intensive Farming and Welfare Regarding Anti-Predator Behavior of Chukar Partridges (*Alectoris chukar*)

Muhammad Bilal

Abstract

Gamebird farming is an emerging industry in Pakistan. Nowadays, the production of large amounts of gamebirds used for restocking purposes is an inescapable prerequisite to compensate for the harvest of wild stocks. The present study aims to delineate the welfare of one of the popular gamebirds Chukar partridge (*Alectoris chukar*, Phasianidae) during intensive farming at the two local game farms. The welfare in terms of anti-predator (AP) behavior was assessed. I analyzed the behavior by arranging AP behavioral assays with a flight initiation test, flight initiation distance, predator test, novel object test, escape test, and flight angles. Specifically, the birds of prey and mammalian predators stimulated the AP behavior in the Chukar partridges. The behavioral assays showed that the Chukar partridge had a clear inclination to escape from predators and can survive if they are to be released into the wild. The initiation of flight was triggered by both avian and mammalian predators indicative of likely good survival chances of these birds. However, it is inferred that restocking and reintroduction of Chukar populations cannot be separated from the adoption of sound welfare programs during intensive rearing.

Keywords: anti-predator behavior, chukar partridge, gamebirds, hunting, intensive farming, reintroduction, welfare

1. Introduction

The hunting of gamebirds for both recreational and food items dates back to ancient times. In some cases, this practice ended up depleting the wild stocks to a considerable extent [1], especially over the last century [2]. To counterbalance this phenomenon, captive breeding for restocking purposes has spread throughout the world. Farming practices to produce, rear, and release game birds improved thanks to the provision of human interventions and care. In this context, the viability of captive-raised birds in the wild is an essential prerequisite.

When released, captive birds face a sudden change in their habitat [3]. They are first provided intense human interventional care, and then, once released, exposed to no or minimal human interventions. Nevertheless, intensively reared birds are of compromised quality in terms of wild behavior and instincts, flight distances and flight speeds, which progressively increase their mortality after release [4–7]. However, less stressed and better-raised birds with stimulated wild instincts for environmental pressures can overcome the associated challenges.

The intensively farmed birds are released for restocking or reintroduction [8–10], but those released for hunting purposes, numbering several million per year, largely exceed the others [9, 11]. Galliformes (especially partridges) account for about 70% of all hunted species [12]. These gallinaceous gamebirds include socioeconomically relevant species such as the Chukar partridge (*Alectoris chukar*), the Red-legged partridge (*A. rufa*), and the Rock partridge (*A. graeca*). These species are appreciated not only for their meat but also for the beauty of their ornamentation.

The Chukar partridge is a medium-sized, diurnal, gregarious, aggressive, and one of the most commonly introduced gamebirds worldwide [13]. It is distributed from southeastern Europe eastwards across the Middle East and Central Asia to Manchuria in the north and Nepal in the south. In Pakistan, it is found from 400 m above sea level (asl) in the Salt Range of Pakistan to 4000 m asl in the Western Himalayas. It is regarded as the national bird of Pakistan, where is vernacularly known as "Chakoor" or Chakor", as well as "Chand Chakor" or "Chan Chakor", due to the appearance of the black line as a crescent on the lateral sides of the face. Now in Pakistan, chukar is widely reared for recreational hunting and reintroduction purposes. However, it is still poorly studied in captivity [14]. It can be reared efficiently for meat production and the food, once curated, is distributed to consumers [15].

The species is indeed exposed to several threats like habitat degradation, and reduction due to human encroachment, hunting, poaching of eggs, chicks, and adults that are used for fighting [16]. The correlation between the success of breeding and weather patterns largely determines its population size [17]. Although the local chukar population in Pakistan seems unaffected by anthropogenic pressures, actually its consistency is dwindling [14]. This chapter is of prime importance due to the focus on the welfare of chukar partridge during intensive farming for reintroduction, meat, and game purposes.

2. Concept of anti-predator behavior

Captive breeding has a prominent effect on the well-being of gamebirds that are exposed to major alterations in their behaviors [18]. Fear of birds, run-down fostering, and motherly care was found to be associated with poor survival skills in the wild. Thereby, there is an increasing concern about natural and semi-natural rearing methods that may guarantee more successful releasing programs [19]. The altered behaviors are partially attributed to the absence of parental care during rearing and lack exposure to predators [20]. Accommodating these drawbacks is costly in terms of both labor and capital [7].

Predators can impact the costs associated with gamebird production in both direct (i.e., predation) and indirect (i.e., disturbance of reproductive birds) ways [8]. The economic losses suffered by the game industry due to predators may reach the order of millions of dollars to the game industry. The anti-predator (AP) training is an economical treatment for developing an early fear practice in chicks that sequel in improved predator avoidance [21]. If AP training is provided during an early age, it can be beneficial in terms of survival in the wild [11].

Many empirical approaches for stimulating and shaping AP behavior can be found in the literature [22–24] and are generally grouped in the following broad categories: (1) The flight initiation (FI) test aims at stimulating a flight response against a human intruder and is quantified by the distance, defined as the minimum distance for eliciting the flight response in birds; (2) The novel object (NO) tests simulate a happenstance with a novel item, usually associated with a food resource [25]; (3) The emergence/escape tests (ET) estimate the propensity of wandering into an unfamiliar or potentially dangerous environment from an instinctively harmless location [26]; Finally, (4) the predator exposure tests present the predator in a controlled manner to elicit its scrutiny or alarm calls under predation danger [27].

On the other hand, boldness - the exploration of the intimate habitat for food or other resources [24]—is usually assessed in conjunction with the threatening stimuli associated with the resources being utilized [24, 28]. In pheasants, boldness is described in terms of their exploration of food and novel objects. Training and application of the aforementioned AP tests can produce near-to-wild behavior in game birds. The early life of chicks in captivity is crucial for the development of such behaviors. Some training can induce stress in birds, but in the long term, these are beneficial [3], assuring the bird's survival in the wild once released.

As validated for other wild species, the AP training should be an essential part and parcel of the captive management of the species' restocking or reintroduction process [29]. Several authors called for the need of carrying out more AP behavior assessments for game birds in captivity (e.g., [2]). Exposure to predators can be potentially beneficial to captive-reared game birds destined to be released. However, the feasibility of providing AP training stimuli should be evaluated carefully. In fact, the consequences of exposure to living predators are still unknown.

When game birds are reared either for conservation, reintroduction, hunting, or meat purposes, it is mandatory to ensure the overall welfare of the birds. This study will ensure welfare through getting insights on the building-up of appropriate wild instincts and behavior in intensively reared Chukars and exploring the potential of captive breeding for future release programs to restock wild Chukar populations. This chapter also aimed to provide guidelines to produce gamebird still maintaining wild behavior through ethological experiments.

3. Study design and pheasantries

This prospective, explanatory and descriptive study aimed to explore current trends in the welfare and management of avian game farming in Pakistan was carried out from May 2020 to August 2021. Two avian game breeding farms, namely Sazgar Wildlife Breeding Farm (SWBF), district Lahore and Padhri Game Farm (PGF) under Padhri Private Game Reserve (PGR), district Jhelum, were chosen as study areas. PGR area is characterized by a subtropical humid climate. SWBF was established under the Punjab Wildlife Act 1973 in 2018. The rearing facility is shown in **Figure 1**. SWBF is located in tehsil Raiwind, district Lahore. Lahore is the capital of the Punjab province and the second-most populous city of Pakistan. It has an altitude range from 200 m to 220 m asl [30].

Figure 1.
Flying pen at SWBF with the characteristic vegetation.

3.1 Experimental birds & ethical consideration

A total of 754 chicks of Chukar partridge were reared intensively at both of the aforementioned breeding facilities. These chicks were reared as per the conservative rearing system for poultry following [31]. The evenness in the management of the birds was ensured. The chicks were offered the typical starter poultry feed, which changed into the grower crumble feed after 4 weeks. Both feed and potable water were provided *ad libitum*. Adequate light was provided during the study periods. Overall, the well-being of both husbandry and management was upheld. At the age of 5–8 weeks, the transportation of birds to the flying pens was started. All the birds were transported in batches of 100 individuals per double case poultry transporting modules. The birds were captured randomly with the aid of a hand net and manumitted in the flying pens for various observations and tests. The welfare of birds during capturing and handling was ensured.

During the present study, no bird was exposed to undue harm or stress. The ethical guidelines for the use of animals in the research studies by the International Society for Applied Ethology [32] were followed. Exposure to natural predators is considered normal and part of the natural niche. The AP training with the live predators can induce temporary stress in the birds [3], but in the long term, it is beneficial. All the birds were captured by hand-net and at the twilight time during the cool. The transporting modules were of proper size and dimensions for the birds under study. Due consideration was given to the time of retention in the modules to keep stress minimum. After the tests, birds were replenished with feed and water (with appropriate anti-stress medicines and Vitamin C) to keep the effect of the tests minimal. All of the birds were observed for any abnormal post-test behavior.

4. Behavioral assessment during intensive rearing

To our knowledge, this is the first comprehensive work providing results of all AP behavioral assays in gallinaceous birds to natural predators in non-contrived settings. For assessing AP behavior, I used the flight initiation test, predator test, novel object test, emergence test, and restraint test [10, 24, 31, 33, 34]. The tests were modified as per the requirements and nature of the Chukars. No birds were exposed to predators during breeding and farming as usually done in intensive farming. In this way, I aimed to assess the significant AP behavior in coveys exposed to the predators in the release pens. Different sample sizes reduced the chances of bias in behavioral monitoring.

4.1 Flight initiation test

The FI test was carried out to find the flight initiation distance (FID) for human intrusion in captivity. The flight initiation behavior was assessed individually by an observer walking toward the flying pen. Firstly, two observers (a volunteer and the author) started walking toward the pen while observing the birds. The recording of observations was started from a distance of 20 meters. Secondly, the observer walked along all sides of the pen. The minimum distance between the enclosure and the observer was 1.5 feet. Meanwhile, the different behaviors of Chukars during the intrusion and stimulation were also assessed and recorded. The FI response was characterized in three cohorts, namely, (1) Weak response: birds slightly changed their position or just walked; (2) Moderate response: Chukars took a short flight from the observer and disturbances; (3) High response: birds flew from one side of the flying pen to the other to escape the disturbance.

A total of 20 observations were carried out on 230 subjects to obtain results on the FI of the Chukar partridges (see Appendix 1). During data collection for the FI test, the sample size was kept at a minimum of $n = 5$ to a maximum of $n = 21$. The mean sample size was 11.5 ± 1.18. These samples were collected from the birds found very near to the mesh wire. I categorized flight initiation responses into weak, medium, and high responses. The maximum number of birds showed high responses ($n = 89$), followed by medium ($n = 71$) and weak ($n = 70$) responses. The results showed that weak and high FI responses are associated with higher variance in the response. The Kruskal-Wallis test showed no significant differences ($p = 0.1817$) between the medians of all three behaviors. High responses were normally distributed as opposed to the weak and medium responses, while medium responses were more skewed (i.e., more abnormally distributed).

4.2 Flight initiation distance FID

Flight Initiation Distance (FID) data collection was carried out independently of the FI test. During the observations for FID, the intrusion distance for Chukars was noted. It is the distance between the human and bird right before the initiation of flight. The bird perceives an intruder (human) as a danger. The bird takes flight when the minimum distance is approached. An observer approached the birds of the flying pens from 10 m outside. When the bird took flight, the distance between the observer and the bird was measured using a measuring tape. A total of 278 observations were recorded. The response categories were divided into 5 cohorts.

Distance cohorts (m)	Frequency	
	Number (n)	Percentage (%)
0–2	30	10.79
2–4	108	38.85
4–6	65	23.38
6–8	59	21.22
8–10	16	5.76
Total	278	100

Table 1.
Showing the distance cohorts and frequencies (numbers and percentages) of FID in those distance cohorts.

The observation was discarded if the bird being intruded did not take flight but just changed its position.

For finding the flight initiation distance of Chukar partridge, a total of 278 observations were recorded. The frequencies of these observations were divided into 5 cohorts (**Table 1**). I found a mean distance of 5.34 ± 0.4 and a variance of 4.55. The minimum and maximum FID recorded were 1.6 m and 9.93 m, respectively. Most of the birds showed FID between 2 m to 4 m (38.85%) followed by 4 m to 6 m (23.38%). The maximum FID was shown by 16 birds (5.76%). Results also indicate that experimental birds have a high FI response in the case of humans. This is beneficial as the trained birds cannot be approached, trapped, or captured after release to the wild. Zaccaroni [21] used the human stimuli for eliciting fights in Rock partridge. That study found more distant escape responses as compared to the non-human intervened birds. Fernández-Juricic et al. [35] described longer FIDs in the species with larger groups or living in flocks, which makes sense based on the more conscious and sensitive birds at the edges or near the disturbances. Hingee and Magrath [36] further elaborated the phenomenon, finding that flight in some birds can cause other less-evoked birds to fly away.

4.3 Predator test (PT)

For Predator Test (PT), the ex-post-facto design in a semi-contrived manner was followed. The natural prevalence and exposure of both avian and mammalian predators were the stimulus for the PT. The AP behavior against raptors was studied during the daytime. Mammalian predators were active at night. The effect of their presence on the birds was assessed. The same types of variables or parameters were used for both types of predators. The behaviors of the birds were also filmed for strengthening the field observations. I noted the behavior of birds until the resuming of the normal activities (feeding or resting) of half of the birds being observed after the predator's appearance. I observed and recorded typical AP behaviors for the birds *viz,* vigilance, crouching, TI and escape. These parameters were characterized as (1) Vigilance: if the birds showed any sign of activeness i.e., outstretched neck with support on front toes, slow staggering gait and actively searching intimate environment; (2) Crouching: this freezing behavior was demarcated by the reduction in the movement or just squatting frequently; (3) Tonic immobility (TI): the persistent hunker position for >30s; and (4) Escape: walking and running, also including flying attempts.

4.3.1 For avian predators

For assessing AP behavior against avian predators, a total of 29 observations were recorded in 515 birds. I noted observations for no more than n = 30 birds. On the other hand, the least sample size was n = 5 birds. Descriptive statistics (see Appendix 2) shows that the mean sample size is 17.75 ± 1.4. The maximum number of birds were found vigilant (n = 18) followed by crouching (n = 15), TI (n = 8), and Escape (n = 6). It can be depicted that crouching and vigilant responses were abundant among the birds. The Mann–Whitney pairwise test shows that there are significant differences in the means of all the behaviors except crouching and vigilance; TI and escape responses. The data also show that 6.65 ± 0.8, 6.75 ± 0.7, 2.44 ± 0.4, and 1.89 ± 0.3 are the means for vigilance, crouching, TI, and escape behaviors for the avian predators, respectively. The highest responses are observed the crouching and vigilance (**Figure 2**). This study evidenced that aerial predators can cause more crouching than vigilance, TI, and escape responses. There were slight differences between crouching and vigilance behaviors. Previous research supported the same results in free-living [37] and captive-reared game birds [38]. Both studies presented those partridges mainly crouched in response to aerial predators. The AP responses are usually accompanied by species-specific alarm calls [39].

4.3.2 For mammalian predators

To assess the behavior of the Chukar partridge upon the encounter with terrestrial predators, I carried out a total of 14 observations on 168 birds. The exposure was non-contrived; in other words, I did not have any impact on their encounter with predators. The results show that escape response was the most frequently recorded (n = 118) in the terrestrial predator case, followed by vigilance (n = 26) and crouching and TI (n = 8 each) (Appendix 3). These results are represented graphically (**Figure 3**). The means for all of the categories of behavior are significantly different ($p < 0.05$). Moreover, Mann–Whitney pairwise test showed significant differences in the means of escape and vigilance, TI and escape. Moreover, there are no significant differences in the means of TI and crouching; TI and vigilance.

Undoubtedly, this study also cleared that escape reaction was the most prevalent behavior in the case of terrestrial predators (see Appendix 3). The least prevalent responses were vigilance, crouching, and TI. By contrast, [38] argued that vigilance was the most common reaction for the exposure to terrestrial predators. It indicated

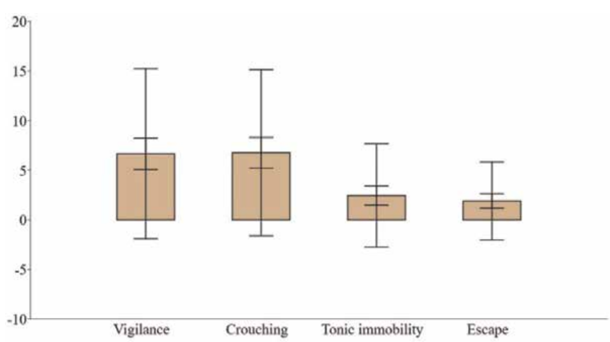

Figure 2.
Graph showing bar chart with standard error (SD) and deviations in the plots at 95% confidence interval for PT against the avian/aerial predators.

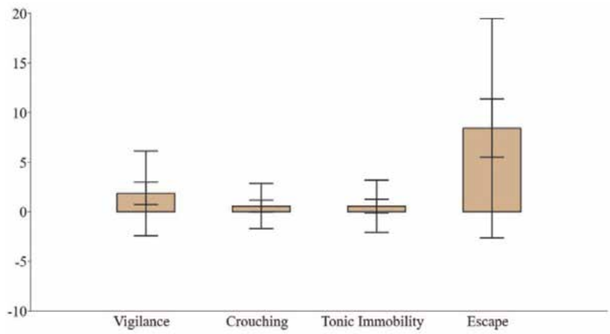

Figure 3.
Graph showing a bar chart with SD and deviations in the plots at 95% confidence interval for PT against the mammalian/terrestrial predators.

the good response to the terrestrial predator by the present birds. Partridges respond differently to different predators [39]. For example, common behavior against terrestrial predators is vigilance, for easy detection and to avoid attack on them. Alternatively, against avian predators the combination of camouflage, TI and crouching can be effective. Interestingly, it was also observed that birds resumed their normal maintenance activities promptly after the exposure to the predators.

The present study had strengths over previous ones for exposure of living predators in de facto design and vast space use. The limitations of the previous results can be attributed to their small-sized experimental cages as compared to those used in our study. Earlier studies by [33, 38, 39], were conducted in cages or pens of very small sizes as compared to the present study. As stated by [40], the experimental cages can disrupt normal physiology and illicit the stress response during the initial transference in the cages. The conducting of experiments in smaller enclosures might have affected the results of the previous studies. Furthermore, the use of dummy predators can be beneficial due to: (1) The experiments and exposure can be presented at one's disposal, (2) Birds cannot be attacked by the dummy predators, and (3) The AP responses of Chukar are stronger for the live predators than dummy predators [41]. Even in captivity, the living predators may cause the deaths of birds in captivity. Meanwhile, the exposure can also stimulate short-term AP behavior in conspecifics in the same pen and the long-run survival in the wild. This behavior can be AP calls, alertness, or flying rigorously. In previous studies, the proper wild behavior would not have been shown by the experimental birds.

4.4 Novel object test (NOT)

For the novel object (NO), test birds were not isolated from the flock. A camera was placed both in and alongside the mesh wire to test the neophobia i.e., the fear of new things. The camera was kept on for recording video. When placing the camera, the disturbance to the birds was kept minimal. If more than three birds were disturbed during the placement of the camera, the observation was discarded for analysis. An observer also carried out a behavioral assay from 40 feet away. A pair of Russian Sehfeld Military binoculars was also used to see birds clearly. The test was continued for 5 minutes or until the birds showed the sign of acclimatization to the object. The NO test was dissolved in the following parameters: (1) Alert and Gazing: birds only gazed at the novel object (camera) and stood alert with no motion, (2) Approach: they

Figure 4.
Experimental settings of the NOT with a covey of 7 Chukars (one behind the pillar) standing vigilant against the novel object (camera).

approached the camera, and (3) Escape: they were fearful of the object and flew away. All three categories of the responses were kept mutually exhaustive.

For NOT, a camera was introduced in the pen and different responses to that novel object were observed (**Figure 4**). The NOT was done on a total of 291 birds. The NOT showed that alertness and gazing was the most frequently observed behavior (n = 166). The approach to the introduced novel object and escape responses were on second (n = 90) and third (n = 35) numbers in frequency. The mean value for the alert and the gazing response was 6.64 ± 0.58, for approach 3.6 ± 0.49. The median value for escape was 1. The ANOVA and Mann–Whitney pairwise test showed that there are statistically significant differences ($p < 0.05$) present in the means of all the behaviors associated with NOT. The descriptive results have been tabulated in Appendix 4.

The briskness in the NOT shows that birds were aware of the danger associated with the introduced object. Very low number of birds perceived the NO as a real danger and escaped, while others approached the object to explore it. Such exploration in the wild would result in the death or trapping of the birds. Furthermore, the birds were not isolated from the covey, so definitely proper behavior against NO is crucial. When birds were in a group, they approached the NO massively (Appendix 4). This finding is in contrast with a study by [42], who found that ravens show neophobia (phobia of new objects) when they are in groups. In other words, they approached the NO more quickly when alone as compared with when they are with conspecifics. Here, partridges preferred to escape when they were alone or in a small covey.

4.5 Escape/emergence test (ET)

ET was done during the transportation of birds into the flying or releasing pens. Transporting modules (80 cm length x 60 cm width x 40 cm height) made of wood were used. A single module has two compartments, each 20 cm in height. All other sides of the module had meshed wire, except the roof and floor sides. Birds could see outside from the sides of the modules. Birds were captured from the training pen using a hand net and kept in the poultry module for 5 minutes for acclimatization. Then birds were carried to the flying pen for liberation. The doors of the modules were opened without disturbance and observers stood as far as possible. The emergence observations were carried out on all birds for 3 minutes. After 3 minutes the test was terminated, and the remaining birds were deemed to show no escape reaction. The escape behavior was divided into three categories: (1) Immobile: after opening the door of transporting module, birds remain inactive or not tried to escape within 3 minutes of the test; (2) Walking: it was an escape response of relatively lower intensity, birds just walked out of the module briskly; and (3) Flying: birds emerged under 3 minutes and took a flight to escape of the module.

ET was performed during releasing of birds to the releasing pen. The escaping pattern of birds from the transporting modules was noted. Results showed that the maximum number of birds (n = 360) did not bother to escape out of the module and remained immobile (24.0 ± 2.9) m (see Appendix 5). The second most frequent response (n = 278) was the flying response (18.53 ± 2.4). It is followed by the least frequent (n = 112) walking response (7.46 ± 1.0). A minimum of 5 birds were found immobile in contrast to 0 and 2 for walking and flying in a single batch. Tukey's pairwise test confirmed the significant comparison between walking and immobile and flying and walking categories. I found no statistically significant differences between the means of immobile and flying categories (p = 0.2123). It is also clear that most of the birds (n = 270) did not try to escape from the boxes at all. The escape from the boxes simulates the potential danger in the intimate environment or attack on the conspecifics. Rather, the majority of the birds preferred to stay motionless in the boxes, while a few others preferred to take flight in a possible perilous situation. The birds flew with a short sound of "keer" [39]. The calls by the escaped birds might elicit stimuli in the birds in boxes for the potential perilous situation outside. Despite this, a work by [43] also found that Chukars did not struggle to escape during the experiment since they showed habituation to the study apparatus.

4.6 Flight angles (FA) test

The birds which showed the flight escape reaction were also subjected to the flight angles test. Flight angles are a proxy of the flight intensity during the escape. The intensity of the flight taken was observed and noted by two independent observers. For this purpose, flight reaction was categorized into three cohorts: (1) Strong flight: the bird took a flight of nearly ≤90° angle, (2) Medium flight: an escape flight of ≤45° angle, (3) Weak flight: the flight of ≤30°. An angle meter was used to measure the angles of the flights. During the flying escape of n = 278 birds, I noted the flight angles. I found that medium flights were the most common mode of escape from the boxes. The mean number of birds for strong flights, medium and weak were 6.26 ± 1.0, 9.2 ± 1.8, and 3.06 ± 0.8, respectively. Out of every batch of maximum strong flights, medium, and weak flights were 15, 24, and 9. Strong flights indicate good escape behavior. The maximum number of birds that took a flight was recorded as n = 38. On the other hand, two were the minimum birds that took flight from any batch. Tukey's pairwise test showed that there was a significant difference between weak and medium flights (p = 0.005). Nevertheless, Mann–Whitney pairwise test showed no statistically significant differences between strong and weak flights, or between medium and strong flights. The descriptive collected data is shown in Appendix 6. Without training, most flights of the birds were in the category of medium ≤45° flight, which is enough to escape a terrestrial predator, but not for avoiding an aerial predator.

Nevertheless, in this study all the birds were naïve to the AP behavior; no AP trainings were provided [38] showed that if red-legged partridge were trained for AP behaviors, they showed a stronger and prolonged response as compared to the inept birds. Present results are not in compliance with this statement, as no AP training was provided before the collection of data. The observed AP behavior cannot be expected to closely overlap with that of the wild conspecifics [33]. If birds were sourced from the wild, there would be different results for AP response. On the other hand, the present study clearly showed that even if early-life trainings were not provided to the game birds, they can still show more than good AP behavior. The differences in the behaviors of birds by simulating the predators (previous studies) and using real natural predators (present study) have significant differences in the intensities and types of shown behaviors.

5. Conclusions

The comprehensive work for AP behavioral assays of Chukar partridge reared intensively was divided into flight initiation test, flight initiation distance, predator test for both avian and mammalian predators, novel object test, escape test, and flight angles. The behavioral assays showed that the Chukar partridge were having the instinct to be released in the wild for restocking and reintroduction purposes. The predators found in the areas have shaped the AP behavior of the birds. The predator tests in the non-contrived settings for the avian species showed that crouching and vigilant responses were the most abundant among the birds. However, the escape response was the highest against the mammalian predators. This study confirms the positive results of the AP tracings via exposure to natural predators in an uncontrolled environment.

For finding the flight initiation distance of Chukar partridge the frequencies of the observations were divided into 5 cohorts. I found a mean distance of 5.34 ± 0.4, while the variance was 4.55. The minimum and maximum FID recorded were 1.6 m and 9.93 m, respectively. Most of the birds showed FID between 2 and 4 m (38.85%). The NOT on 291 subjects showed that alertness and gazing were the most frequently shown behavior. It was observed that the maximum number of birds did not escape from the module and remained immobile. During the flying escape of birds, the flight angles were also noted. The most common intensity of escape from the boxes was medium flight. The FI test showed that the maximum number of birds has high responses followed by medium and weak responses. Both weak and high FI responses showed more variance in the response.

The present study focused on the main issues of the management of the gamebird species in Pakistan, also making a contribution to the improvement of AP behavior primarily in the Chukar partridge reared intensively for both restocking and reintroduction purposes. This study can be replicated on Ring-necked pheasants, commonly reared for shooting. Future works should focus on the following recommendations.

1. The cost-effective training for the improvement of AP behavior should be a priority for game breeders. The pre-release assessment of AP behavior should be upheld.

2. The natural predators of gamebirds should be present at farm locations to elicit predator-specific responses. The new breeders and game bird-providing companies should keep this point in consideration when preparing for the breeding.

Acknowledgements

The author acknowledges the technical support and provision of equipment for this study by HE. Ch.. Suleyman Khan Warraich.

Appendix 1. Sample sizes of birds and their FI responses against the human disturbances

No.	Sample size	Weak	%	Medium	%	High	%
1	18	12	66.67	4	22.22	2	11.11
2	7	4	57.14	1	14.29	2	28.57
3	21	16	76.19	3	14.29	2	9.52
4	9	4	44.44	3	33.33	2	22.22
5	5	2	40.00	2	40.00	1	20.00
6	14	3	21.43	6	42.86	5	35.71
7	11	6	54.55	3	27.27	2	18.18
8	7	2	28.57	3	42.86	2	28.57
9	19	2	10.53	5	26.32	12	63.16
10	13	6	46.15	2	15.38	5	38.46
11	5	1	20.00	3	60.00	1	20.00
12	9	1	11.11	5	55.56	3	33.33
13	16	3	18.75	7	43.75	6	37.50
14	6	1	16.67	2	33.33	3	50.00
15	18	2	11.11	5	27.78	11	61.11
16	5	1	20.00	1	20.00	3	60.00
17	17	1	5.88	4	23.53	12	70.59
18	13	1	7.69	5	38.46	7	53.85
19	11	1	9.09	4	36.36	6	54.55
20	6	1	16.67	3	22.22	2	33.33
Total	230	70		71		89	
Variance		15.84		2.57		12.68	

Appendix 2. Four categories of responses namely vigilance, crouching, TI and escape for predator test against the avian predators

No.	Sample	Vigilance		Crouching		Tonic immobility		Escape	
		n	%	n	%	n	%	n	%
1	22	3	13.64	11	50.00	4	18.18	4	18.18
2	20	4	20.00	8	40.00	5	25.00	3	15.00

No.	Sample	Vigilance		Crouching		Tonic immobility		Escape	
		n	%	n	%	n	%	n	%
3	27	18	66.67	3	11.11	0	0.00	6	22.22
4	7	3	42.86	3	42.86	0	0.00	1	14.29
5	29	9	31.03	11	37.93	5	17.24	4	13.79
6	22	2	9.09	13	59.09	7	31.82	0	0.00
7	17	5	29.41	12	70.59	0	0.00	0	0.00
8	16	7	43.75	7	43.75	2	12.50	0	0.00
9	29	15	51.72	4	13.79	4	13.79	6	20.69
10	12	4	33.33	3	25.00	3	25.00	2	16.67
11	24	9	37.50	12	50.00	1	4.17	2	8.33
12	27	7	25.93	15	55.56	0	0.00	5	18.52
13	6	1	16.67	3	50.00	0	0.00	2	33.33
14	25	14	56.00	0	0.00	8	32.00	3	12.00
15	23	10	43.48	5	21.74	7	30.43	1	4.35
16	5	2	40.00	1	20.00	1	20.00	1	20.00
17	17	11	64.71	4	23.53	0	0.00	2	11.76
18	22	4	18.18	11	50.00	7	31.82	0	0.00
19	19	9	47.37	5	26.32	2	10.53	3	15.79
20	5	2	40.00	3	60.00	0	0.00	0	0.00
21	15	3	20.00	9	60.00	3	20.00	0	0.00
22	25	9	36.00	9	36.00	3	12.00	4	16.00
23	13	2	15.38	9	69.23	2	15.38	0	0.00
24	8	7	87.50	1	12.50	0	0.00	0	0.00
25	24	10	41.67	13	54.17	0	0.00	1	4.17
26	16	10	62.50	6	37.50	0	0.00	0	0.00
27	6	2	33.33	3	50.00	1	16.67	0	0.00
28	20	6	30.00	3	15.00	6	30.00	5	25.00
29	14	5	35.71	9	64.29	0	0.00	0	0.00
Total	515	193		196		71		55	

Appendix 3. Comparison of four categories of the AP responses namely vigilance, crouching, TI and escape for predator test against the mammalian predators

No.	Sample	Vigilance		Crouching		Tonic Immobility		Escape	
		n	%	N	%	N	%	N	%
1	14	7	50.00	0	0.00	5	35.71	2	14.29

No.	Sample	Vigilance		Crouching		Tonic Immobility		Escape	
		n	%	N	%	N	%	N	%
2	4	0	0.00	0	0.00	0	0.00	4	100.00
3	14	5	35.71	2	14.29	0	0.00	5	35.71
4	15	3	20.00	0	0.00	0	0.00	12	80.00
5	4	0	0.00	0	0.00	0	0.00	4	100.00
6	6	1	16.67	0	0.00	1	16.67	4	66.67
7	22	0	0.00	3	13.64	0	0.00	16	72.73
8	11	3	27.27	0	0.00	1	9.09	7	63.64
9	15	0	0.00	0	0.00	0	0.00	15	100.00
10	7	2	28.57	0	0.00	0	0.00	5	71.43
11	22	0	0.00	3	13.64	0	0.00	16	72.73
12	7	3	42.86	0	0.00	0	0.00	4	57.14
13	18	0	0.00	0	0.00	0	0.00	18	100.00
14	9	2	22.22	0	0.00	1	11.11	6	66.67
Total	168	26		8		8		118	

Appendix 4. Results of NOT for the three responses namely alert & gazing, approaching and escape for the introduced novel object in the pen

No.	Samples	Alert & Gazing		Approach		Escape	
		n	%	n	%	n	%
1	16	10	62.5	4	25	2	12.5
2	11	5	45.45	5	45.45	1	9.09
3	7	3	42.86	3	42.86	1	14.29
4	13	8	61.54	4	30.77	1	7.69
5	15	6	40	5	33.33	4	26.67
6	6	5	83.33	1	16.67	0	0
7	4	1	25	2	50	1	25
8	9	9	100	0	0	0	0
9	13	6	46.15	5	38.46	2	15.38
10	12	9	75	3	25	0	0
11	20	10	50	7	35	3	15
12	7	3	42.86	3	42.86	1	14.29
13	10	6	60	3	30	1	10
14	18	9	50	7	38.89	2	11.11

No.	Samples	Alert & Gazing		Approach		Escape	
		n	%	n	%	n	%
15	13	10	76.92	2	15.38	1	7.69
16	8	7	87.5	1	12.5	0	0
17	18	12	66.67	5	27.78	1	5.56
18	12	9	75	3	25	0	0
19	18	10	55.56	4	22.22	4	22.22
20	10	7	70	2	20	1	10
21	17	6	35.29	9	52.94	2	11.76
22	3	3	100	0	0	0	0
23	6	5	83.33	1	16.67	0	0
24	20	5	25	9	45	6	30
25	5	2	40	2	40	1	20
Total	291	166		90		35	

Appendix 5. Data for the ET and different categories of responses namely immobile, walking and flying for the escaping behavior out of the transporting module

No.	Samples	Immobile		Walking		Flying	
		n	%	N	%	n	%
1	50	28	56	5	10	17	34
2	50	32	64	7	14	11	22
3	50	8	16	13	26	29	58
4	50	30	60	14	28	6	12
5	50	16	32	13	26	21	42
6	50	25	50	9	18	16	32
7	50	48	96	0	0	2	4
8	50	22	44	8	16	20	40
9	50	42	84	2	4	6	12
10	50	25	50	3	6	22	44
11	50	16	32	9	18	25	50
12	50	22	44	5	10	23	46
13	50	5	10	7	14	38	76
14	50	20	40	5	10	25	50
15	50	21	42	12	24	17	34
Total	750	360		112		278	

Appendix 6. Results of the flight angles data and associated flight intensity angles namely, strong ≤90°, medium ≤45° and weak ≤30° when took flight out of the transporting modules

No.	Samples	Strong ≤90°		Medium ≤45°		Weak ≤30°	
		n	%	N	%	n	%
1	17	11	64.71	5	29.41	1	5.88
2	11	3	27.27	7	63.64	1	9.09
3	29	12	41.38	16	55.17	1	3.45
4	6	2	33.33	3	50.00	1	16.67
5	21	4	19.05	9	42.86	8	38.10
6	16	7	43.75	5	31.25	4	25.00
7	2	1	50.00	1	50.00	0	0.00
8	20	7	35.00	8	40.00	5	25.00
9	6	3	50.00	3	50.00	0	0.00
10	22	9	40.91	11	50.00	2	9.09
11	25	1	4.00	24	96.00	0	0.00
12	23	8	34.78	10	43.48	5	21.74
13	38	6	15.79	23	60.53	9	23.68
14	25	15	60.00	2	8.00	8	32.00
15	17	5	29.41	11	64.71	1	5.88
Total	278	94		138		46	

Author details

Muhammad Bilal
Department of Biology, Virtual University of Pakistan, Rawalpindi, Pakistan

*Address all correspondence to: bilalvu4@gmail.com

References

[1] Hanane S, Magri N. Post-release habitat utilisation by Francolinus bicalcaratus ayesha, a critically endangered subspecies endemic to Morocco: Implications for optimising future release programmes. Bird Conservation International. 2016;**26**: 323-336

[2] Tapper SC. A Question of Balance: Game Animals and Their Role in the British Countryside. Fordingbridge: Game Conservancy Trust; 1999

[3] Madden JR, Santilli F, Whiteside M. The welfare of game birds destined for release into the wild: A balance between early life care and preparation for future natural hazards. Animal Welfare. 2020; **29**:1-18. DOI: 10.7120/09627286.29.1.001

[4] Pérez JA. Use of radiotracking techniques to study a summer repopulation with red-legged partridge (Alectoris rufa) chicks. Poultry Science. 2004;**83**:882-888

[5] Alonso ME. Study of survival, dispersal and home range of autumnreleased red-legged partridges (*Alectoris rufa*). British Poultry Science. 2005;**46**:401-406

[6] Parish DM. The fate of released captive-reared grey partridges Perdix: Implications for reintroduction programmes. Wildlife Biology. 2007;**13**: 140-149

[7] Buner F, Browne S, Aebischer N. Experimental assessment of release methods for the reestablishment of a red-listed galliform, the grey partridge (Perdix perdix). Biological Conservation. 2011;**144**(1):593-601. DOI: 10.1016/j.biocon.2010.10.017

[8] Park K, Graham K, Calladine J, Wernham C. Impacts of birds of prey on gamebirds in UK: A review. Ibis. 2008; **150**(s1):9-26

[9] Sokos CK, Birtsas PK, Tsachalidis EP. The aims of galliforms release and choice of techniques. Wildlife Biology. 2008;**14**: 412-422. DOI: 10.2981/0909-6396-14.4.412

[10] Santilli F, Bagliacca M. Fear and behavior of young pheasants reared with or without parent figure. Avian Biology Research. 2019;**12**(1):23-27. DOI: 10.1177/1758155919826765

[11] Gaudioso VR, Sánchez-García C, Pérez JA, Rodríguez PL, Armenteros JA, Alonso ME. Does early antipredator training increase the suitability of captive red-legged partridges (*Alectoris rufa*) for releasing? Poultry Science. 2011;**90**:1900-1908. DOI: 10.3382/ps.2011-01430

[12] Martinez J, Vinuela J, Villafuerte R. Socio-Economic and Cultural Aspects of Gamebird Hunting. Finland: University of Turku; 2002

[13] Smith AM, Cropper WP, Moulton MP. A quantitative assessment of site-level factors in influencing chukar (Alectoris chukar) introduction outcomes. PeerJ. 2021;**9**:e11280. DOI: 10.7717/peerj.11280

[14] Khattak RH, Xin Z. Precise and accurate behavioral monitoring of captive chukar (Alectoris chukar) by using the advanced digital ethogram: A small scale pilot study. Brazilian Journal of Biology. 2021;**83**:e246053. DOI: 10.1590/1519-6984.246053

[15] Tyasi TL, Putra WP. The growth curve of body weight in chukar patridges (Alectoris chukar) based on Richards

mathematical model. In: The 3rd International Conference on Biosciences and Medical Engineering. Malaysia: Universiti Teknologi Malaysia; 2021. Available online: https://www.utm.my/icbme/

[16] Ahmad B, Nabi G, Hayat M, Muhammad J. Habitat study of chukar partrudge (Alectoris chukar) in district Malakand: A case study of town Thana, Pakistan. Journal of Biodiversity and Environmental Sciences. 2017;**11**(4):11-17

[17] Duarte JM. New data on mortality, home range and dispersal of red-legged partridge (Alectoris rufa) released in a mountain range. European Journal of Wildlife Research. 2010;**57**:675-678. DOI: 10.1007/s10344-010-0467-9

[18] Mason G. Species differences in responses to captivity: Stress, welfare and the comparative method. Trends in Ecology & Evolution. 2010;**25**(12):713-721

[19] Browne SJ, Buner F, Aebisvher NJ. A review of gray partridge restocking in the UK and its implications for the UK biodiversity action plan. National Quail Symposium Proceedings. 2009;**6**(1):40

[20] Rantanen EM. Vigilance, time budgets and predation risk in reintroduced captive-bred grey partridges Perdix. Applied Animal Behavioral Science. 2010;**127**:43-50

[21] Zaccaroni MC. Does an early aversive experience to humans modify antipredator behavior in adult rock partridges? Ethology Ecology and Evolution. 2007;**19**:193-200. DOI: 10.1080/08927014.2007.9522561

[22] Reale D, Reader S, Sol D, McDougall P, Dingemanse N. Integrating animal temperament within ecology and evolution. Biological Reviews. 2007;**82**:291-318

[23] Carter A, Feeney W, Marshall H, Cowlishaw G, Heinsohn R. Animal personality: What are behavioral ecologists measuring? Biological Reviews. 2012;**88**:465-475

[24] Kerman K, Sievingb KE, Maryc CS, Avery ML. Evaluation of boldness assays and associated behavioral measures in a social parrot, monk parakeet (*Myiopsitta monachus*). Behavior. 2016;**153**: 1817-1838. DOI: 10.1163/1568539X-00003356

[25] Garamszegi L, Rosivall B, Rettenbacher S, Marko G, Zsebok S, Szollosi E. Corticosterone, avoidance of novelty, risk-taking and aggression in a wild bird: No evidence for pleiotropic effects. Ethology. 2012;**118**:621-635

[26] Miller K, Mench J. Is fearfulness a trait that can be measured with behavioral tests? A validation of four fear tests for Japanese quail. Animal Behavior. 2006;**71**:1323-1334

[27] Wilson A, Godin J. Boldness and behavioral syndromes in the bluegill sunfish, Lepomis macrochirus. Behavioral Ecology. 2009;**20**:231-237

[28] Campo J, Dávila S, Gil M, Torres O, Moreno J. Fear and stress measurements in pure and hybrid red-legged partridges. Applied Animal Behavior Science. 2015;**166**:123-130

[29] Shier D, Owings D. Effects of predator training on behavior and post-release survival of captive prairie dogs (Cynomys ludovicianus). Biological Conservation. 2006;**132**:126-135. DOI: 10.1016/j.biocon.2006.03.020

[30] Muhamamd AM, Zhonghua T, Dawood AS, Earl B. Evaluation of local groundwater vulnerability based on DRASTIC index method in Lahore,

Pakistan. Geofísica Internacional. 2015; **54**(1):67-81

[31] Muneer M, Bilal M, Ditta A. A comparative study of some hematological parameters of broiler and indigenous breeds of poultry. SVU-International Journal of Agricultural Sciences. 2021;**3**(4):189-199. DOI: 10.21608/svuijas.2021.99388.1146

[32] Sherwin C, Christiansen S, Duncan I, Erhard H, Lay D, Mench J. Guidelines for the ethical use of animals in applied ethology studies. Applied Animal Behavior Science. 2003;**81**:291-305. DOI: 10.1016/S0168-1591(02)00288-5

[33] Campler M, Jöngren M, Jensen P. Fearfulness in Red junglefowl and domesticated white Leghorn chickens. Behavioral Processes. 2009;**81**:39-43

[34] Davila SG, Cuenca OT, Gil MG, Tolledano-Diaz OA, Castano C, Santiago-Moreno J. Anti-predator behavior in pure and hybrid red-legged partridges. Poultry Science. 2019;**98**: 3085-3092

[35] Fernández-Juricic E, Jimenez MD, Lucas E. Factors affecting intra- and inter-specific variations in the difference between alert distance and flight distances for birds in forested habitats. Canadian Journal of Zoology. 2002;**80**: 1212-1220

[36] Hingee M, Magrath RD. Flights of fear: A mechanical wing whistle sounds the alarm in a flocking bird. Proceedings of the Royal Society of London, Series B: Biological Sciences. 2009;**276**:4173-4179

[37] Buner F. Survival, Habitat Use and Disturbance Behavior of re-Introduced Grey Partridges Perdix L. in an Enhanced Arable Landscape in the Swiss Klettgau. Switzerland: University of Basel and Swiss Ornithological Institute; 2007

[38] González-Redondo P, Delgado-Pertíñez M, Toribio S, Ruiz F, Mena Y, Caravaca F. Characterisation and typification of the red-legged partridge (Alectoris rufa) game farms in Spain. Spanish Journal of Agricultural Research. 2010;**8**(3):624-633. DOI: 10.5424/sjar/2010083-1258

[39] Binazzi R, Zaccaroni M, Nespoli A, Massolo A, Dessi-Fulgheri F. Anti-predator behavior of the red-legged partridge Alectoris rufa (Galliformes: Phasianidae) to simulated terrestrial and aerial predators. The Italian Journal of Zoology. 2011;**78**(1):106-112

[40] Dickens M, Earle K, Romero L. Initial transference of wild birds to captivity alters stress physiology. General and Comparative Endocrinology. 2009;**160**:76-83. DOI: 10.1016/j.ygcen.2008.10.023

[41] Bilal M, Piebeng GNK, Naseer A. The anti-predator behavioral assays of Chukar partridges (Alectoris chukar) against the live predators in captivity. In: The Scientific Days. Maroua: University of Maroua; 2022. p. 121

[42] StoI M, Bugnyar T, Heinrich B, Kotrschal K. Effects of group size on approach to novel objects in raven (*Corvus corax*). Ethology. 2006;**112**: 1079-1088

[43] Chappell MA, Bucher TL. Effects of temperature and altitude on ventilation and gas exchange in chukars (*Alectoris chukar*). Journal of Comparative Physiology B. 1987;**157**:129-136

Chapter 3

A Global Overview of the Intensification of Beef and Dairy Cattle Production Systems

Grace Opadoyin Tona

Abstract

The global livestock agriculture, including the beef and dairy cattle production systems, has undergone several transformations from traditionally less productive into more productive intensive systems. This research work reviews the various tools and techniques that have enhanced the development of more intensive beef and dairy cattle production worldwide. There is advancement from the extensive grazing on rangelands, into the more intensive systems of production under confined housing in the semi-intensive and intensive systems. Several investments would be required in the form of housing, feeding, breeding and genetic improvement, health and animal welfare and policy designs by the low-income livestock farmers, commercial livestock farmers and the larger livestock industries and governmental agencies. The increasing global population made it imperative to seek for more intensive and sustainable systems of beef and dairy cattle production in order to meet the human need for the production of cost-effective animal protein sources in the form of beef and bovine milk.

Keywords: beef and dairy cattle, intensified management systems, genetic improvement, sustainability, production systems

1. Introduction

Beef and dairy cattle play the unique role of providing high quality protein for human consumption from forage and other concentrated feed resources. Globally the cattle sourced protein is the most common, popular, most available, acceptable and affordable by most people in terms of cost per unit weight. The intensive production of beef and milk sourced from cattle through the use of new technologies in livestock agriculture could lead to increased number of animals with greater yield in livestock products such as beef and milk [1]. The enhancing technologies could be in the form of mechanical, biological and chemical tools. The use of larger and faster implements would enable the need for lesser human power to operate and achieve larger land cultivation, leading to higher beef and milk yield. Thereafter, the application of chemical fertilizers and herbicides would increase the rate of forage cultivation and feed production. The authors [1] further outlined that animal drugs and antibiotics use would result to increased animal health, reduction in mortality accompanied by increased beef and milk production under the lower levels of input application. These could thus result into higher profit margin due to lowered cost of production.

The growing human food needs of an expanding human population, the challenges of global climate change have resulted in the need for the development of sustainable ruminant production systems [2]. It has been projected by FAO [3] that the world population would become 9.73 billion by 2050 and 11.2 billion by 2100. Globally, most young people are expected to live in the Sub-Saharan Africa and Asia, and particularly in the rural areas where higher rate of unemployment exist. Thus, there could arise the situation whereby there would be a world population increases, there would be high rate of urbanization by the youth, with the aged people left in the rural agricultural cultivation and production areas. These could make it difficult to meet the requirement for agricultural labour force and socioeconomic structure needed by the rural community to achieve sustainable development goals [3]. A group of researchers [4], reported for beef cattle production while another group [5], reported for dairy cattle production that through the intensification of beef and dairy cattle production systems, genetic selection for either beef or dairy cattle, and the use of modern technologies such as artificial insemination and genomic selection, the beef and dairy cattle industries have more than doubled in their production over past decades. However, there had been the contrasting dramatic reduction in the total number of pure-bred cattle being raised globally either for beef or for milk. There were also observed few unfavourable responses in relation to fertility, health challenges, longevity and environmental sensitivity. Again, as earlier mentioned, large numbers of offsprings from pure-bred dairy cows do enter beef fattening systems as milk-fed veal producing beef cattle [6]. Thus, this article reviews the intensified systems of beef and dairy cattle production worldwide.

2. Beef and dairy cattle production systems

2.1 Beef production systems

It was outlined [7] that the system of commercial beef production may be classified into three general categories namely: (i) A cow-calf division wherein weaned feeder calves were raised for further grazing mainly on pastureland and/or feeding with other concentrate diets. (ii) The back-grounding or stocker phase of production wherein body weight was gained by the recently weaned calves, resulting into feedlot-ready yearlings which also took place mainly on pastureland. (iii) The finishing phase of beef production wherein cattle were fattened for slaughter was carried out mainly intensively under confinement.

The intensive beef production occurs during the finishing stage and also there was the dairy veal production in which beef was produced as a by-product of dairy cows, and these mostly occurred in the developed countries [4]. The researcher [4] reported that in the USA and Europe, there were specialized production, whereby there were mainly beef only, and dairy beef and veal were meant for domestic consumption. In the New Zealand, beef obtained from the dairy industry were produced mainly for export. In the UK, cows raised in the dairying system were increasingly being mated with beef breed sires to upgrade the quantity and quality of beef and veal produced through the use of sexed semen for a particular sex offspring.

2.1.1 Cow-calf (beef) system

The cows and calves were raised under the grazing and rangeland production with strategic nutritional supplementation with concentrate rations and by-products. Their reproductive efficiency could be defined as the weight of calves weaned per cows mated successfully per year. The reproductive rate was thus measured as the percentage of calves weaned relative to the cows mated successfully. It was explained [4] that the maternal genotype was very important and when combined with nutritional and other environmental factors, these go a long way to enhance good reproductive efficiency in beef cattle. Weaned and growing cattle need adequate feed intake from pasture as well as supplements, to ensure rapid growth and improved productivity.

2.1.2 Back-grounding or stocker system

These involved the practice of grazing and foraging of beef cattle in rangelands over large land areas under harsh environmental conditions of rain and sunshine. The grazing and foraging types included continuous, set-stocked, rotational, strip, strategic, cell or time controlled and planned systems [4]. Genotypes that were reported as used under these beef production systems included the *Bos taurus* breeds and crosses, and the Wagyu and Wagyu crosses.

2.1.3 Feedlot and other intensive beef production systems

Feedlots were described as the systems of raising cattle to finishing stage, from an initial liveweight of 280 kg until about 400 kg liveweight. Feedlots were used mainly to fatten cattle when the available pasture was inadequate to meet the nutritional requirement of the animals during the period of drought. In two research outcomes [8, 9], it was observed that feedlot diets in the USA and Australia were usually of high protein levels in excess of the requirement for cattle that had reached the fattening or finisher stage. Feedlot diets were used to maximize feed: gain ratio in order to obtain maximum growth rate. In two groups of researches, [10] for Japanese Wagyu and another group [11] for South Korean Hanwoo cattle, it was mentioned that beef under beef production systems were raised to produce beef animals with optimum levels of intramuscular fat development to achieve highly marbled beef. The Wagyu breed cattle kept on feedlots were fed in groups with high energy diets, which were offered twice or thrice daily, from about 11 months of age until 28–30 months at slaughter with water, mineral salt blocks, salt and diuretic provided *ad libitum* [10].

2.2 Dairy cattle production systems

Dairy cattle could be raised under intensive, semi-intensive or extensive housing systems.

2.2.1 Intensive or confinement system

Most cattle in the USA and Europe were kept in confinement free-stall, tie-stall and dry-lot or bedded pack systems. In these two countries, only few dairy animals were kept on pastureland. Under the intensive system were the tie-stall and the free-stall systems. Various milking parlour types existed, and the free-stall system gave cows more freedom of movement as compared to the tie-stall [12]. In the intensive management, cows that were raised under confinement generally produced more milk than those raised on pasture. These exotic cow breeds produced milk in the range of 7,000–14,000 kg per cow per lactation of around 305 days. They were milked twice or at most thrice daily. Cows were usually fed total mixed ration made up of forages and concentrates separately. The concentrate diets usually consisted of grains, protein sources, minerals and vitamins. All the lactating cows could be grouped and taken orderly into the milking parlour through holding areas and clean cool water was provided *ad-libitum*. Milking parlour types included the low-cost step-up, herringbone, parallel or rotary [12].

2.2.2 The semi-intensive and extensive systems

In South America, Australia, New Zealand and India, dairy cattle were raised under semi-intensive and extensive systems, which were the pasture-intensive systems. These were more commonly used rather than the intensive system.

2.3 Housing of dairy calves

Calves were usually separated from their dams soon after birth and bottle fed with milk or milk replacer (about 4–5 litres/day) twice or thrice daily. Calves were housed in small groups and should have access to clean water *ad-libitum*. Culled calves were usually raised for beef.

2.4 Breeding of dairy cows

Dairy cows were usually bred at 12–16 months of age. They could calve at around 24 months old, and could be milked as heifers for an average of 305 days in lactation. They could then be made to have a dry period of 50–60 days whereby they were not milked (during the third trimester pregnancy period) before they calved, and thereafter to start another reproductive cycle for another lactation period.

3. Genetic improvement programs

In the dairy cattle industry, particularly with the raising of exotic breeds, there had been doubled amounts in milk production globally over the past 50 years (5 decades), while there were declining rates in the dairy cows' population [5]. These were achieved through the intensification of the milk production systems and direct genetic selection for milk yield and other related traits. There were developed the application of modern technologies such as artificial insemination and genomic selection [5]. There had been intensified inbreeding among few well known breeds for high milk production. The positive results of the intensive selection for milk yield however also brought about unfavourable genetic responses such as fertility reduction, adverse effect on the health, longevity and environmental responses sensitivity in the dairy cows. Despite these drawbacks, breeding goals needed to be continued with the focus towards animal welfare, health and longevity, without sacrificing the need to maintain the traits for high milk yield. Achievements were also directed towards breeding for traits that were heritable in terms of their phenotypic expressions. There were also the attainment of long term sustainability goals geared towards organic farming cultures, and pasture-based and mountain-grazing farming systems.

3.1 Breeding for high milk production

3.1.1 Some unfavourable outcomes of breeding for high milk production

Intense selection for high milk production was observed to result in unfavourably correlated responses on other important traits [13] such as conception rate, health (SCC), less control over environmental temperatures, relative humidity and wind speed. There were also more dependence on human feed inputs such as cereals, protein sources and high quality forages. Thus, the observation pointed to the fact that genetic improvement might require more sophisticated approaches.

Some of the developed solutions to these included the incorporation of female fertility traits into breeding programs, improvement in nutrition, health and cows' comfort. Also, dairy farmers on becoming aware of the negative results and the consequences of continuous practice of inbreeding were still seeking mitigation alternatives such as the use of mating software and assistance from extension specialists [13].

3.1.2 Cost effective ways to genetically improve milk production in local dairy cattle breeds

In the developing countries where there were mainly low milk producing local dairy cattle breeds, the cows could continually be upgraded through the application of controlled cross-breeding programs. Local dairy cattle breeds could be crossbred with exotic breeds with the aim of developing new composite breeds. Different selection indices would need to be developed to select animals with higher milk production and that could perform well in terms of animal welfare or fitness, health and longevity under an economically sustainable production system. The cross-breeding of local dairy cattle breeds with exotic dairy cattle breeds such as the Holstein Friesian and Jersey could be improved upon to lead to high milk production, while at the same time there could result improved adaptation to the environment, and the animals also retaining their fitness [5].

In all these research efforts much care need be taken to avoid losses in the achieved genetic diversity traits among the dairy cattle populations [5]. Some other group of researchers [14] pointed out that the refinement of breeding programs to incorporate novel breeding objectives required the development of high-throughput phenotyping technologies such as structural and continuous data recording streams and the investigation of the genetic relationship between novel traits and those that were commonly observed. There could be large scale genome studies, especially genomic predictions and genome-wide association studies, refinement of selection indices to reflect improved knowledge of Biology, new resources of data and changing conditions in the environment. Some of the novel traits in dairy production included health (udder health, hoof health and metabolic disorders), fertility, feed efficiency, methane emissions efficiency, longevity and overall resilience [14].

3.1.3 Innovations and technological breakthroughs in the development of intensive dairy cattle production selection systems

These could be outlined as follows:

i. Conventional genetic selection [15]

The conventional selection was primarily geared towards genetic selection for high-producing dairy cows where milk yield was seen as the main objective towards dairy farming intensification and its sustainability from a single perspective of genetics. Thus milk yield and composition were the main focus for selection in dairy cattle breeding programs and these were achieved in all the leading milk producing dairy cows globally, which included the Ayrshire, Brown Swiss, Guernsey, Holstein, Jersey and Milking Shorthorn [4].

Therefore, high milk yield was often seen as the major solution to address the global challenges of ensuring food security, reducing greenhouse gas (GHG) emissions, reduced cost of milk production, and all these could result into better feed efficiency [16].

ii. Use of conceptual framework with application of standardized environments which employ the equation: P = G + E, where P = phenotype/performance, G = additive genetic merit and E = environmental effect.

iii. Use of advances in nutritional practices and precision management.

iv. Adoption of reproductive techniques such as artificial insemination, embryo transfer and sexed semen.

v. Use of precision health and care management.

vi. The focus on animal health and welfare, and environmental efficiency for management of large waste produced by the cows.

In a practical application in the use of selection indices, some researchers [5] reported that in the USA for example, their selection indices in the dairy cattle industries included such traits as health, SCC (somatic cell counts), live-ability, productive life, feet and legs traits, calving ability and lower usage of antibiotics.

3.2 The intensification of global beef production

Beef provides the largest proportion of ruminant protein meat source world-wide in both the developed and developing countries.

3.2.1 In the USA

Beef production in the USA [17], was reported to be majorly pastoral based particularly under the national cow-calf herd system of production. There were also the use of eighty different beef cattle breeds with the British breeds and their crosses most prevalent. A high income was obtained through beef sales in the U.S.A. These same authors [17] stated that in 2015, about 29 million head of beef were slaughtered in the production of 10.7 million tonnes of beef and the total farm cash receipts total about US$ 88 billion.

The various factors considered during the quality grading of beef by consumers in the USA included the following: colour, marbling level, subcutaneous fat trim and cut thickness. Beef palatability grading by consumers was usually carried out based on the criteria of tenderness, juiciness and flavour [17].

Technological improvements made in the beef industry in the USA included roles in reproduction, feeding and feed processing, animal health, animal productivity including the use of acceptable growth promotants and genomically-enhanced genetic selection and food safety.

The sustainable intensification of beef production in the USA was considered of great importance in order to help meet the rising global demand for lower cost beef protein resource, needed to meet the increasing yearly world-wide population growth rate [17].

3.2.2 Australia beef production

In Australia there were reported to be about 47,000 cattle ranch owners who contributed about 20% of the total value of farm production ($A 12.7billion Gross Value of Production) [17]. The country was one of the world's most efficient producers of cattle, and ranked as the third largest exporter of beef. In 2016–2017 the Australian beef industry had about 25 million heads of cattle. They were identified to produce high quality beef under environmentally sustainable systems of disease-free cattle that followed strict livestock and meat quality regulations and quality assurance systems. Australia was reported to operate under the national beef genetic improvement program called BREEDPLAN which consisted of many temperate and tropically adapted breeds of cattle [17]. These beef cattle breeds were found suitable for different types of agro-climate conditions and were genetically selected based on productivity and market related traits [17]. Australia was known to export beef to various Asian countries including Japan, Korea, China and Indonesia in 1987 to 2009.

4. Sustainable development

4.1 Manure handling and management

Some researchers [1] reported that industrialized beef and dairy cattle production concentrated manure on limited land areas. A small proportion of the cost of production (about 3 %) was usually spent as cost of manure removal in large farms. Manure was usually applied as fertilizer on crop land and most of the manure needed to be well managed so as not to pollute the land surface water and the atmosphere through the volatilization of gases (GHG) and odours.

4.2 Greenhouse gas (GHG) emissions

It was mentioned in a previous study [18] that livestock production such as the production of beef and bovine milk contributed 14.5 % of global GHG emissions. However, since the human population is expected to be increased from 7.2 to 9.6 billion by 2050, there could be increased demand for livestock products. Therefore, the raising of beef and dairy cattle could be found to contribute less in the mitigation (making less severe) of GHG emissions. Another researcher [19] stated that the raising of fewer numbers of more genetically productive breeds of cattle under an intensive system could have positive impact on the mitigation of GHG emissions. Other previous researchers [20] also predicted that methane emissions from domesticated ruminants such as beef and dairy cattle in sub-Saharan Africa could have increased by 40% between 2008 and 2030 due to an increased livestock population. It was however explained [21] that probably the only effective way of methane mitigation in pastoral systems could be through reducing livestock numbers by promoting the intensive livestock production systems.

Another research finding [22], outlined that substantial amounts of carbon could be sequestrated through improvement in the management of grasslands. This would involve the conversion of low degraded cropland or woodland into grasslands. These processes of grazing lands transformation into grasslands could also be enhanced by ensuring reduction in grazing intensities, reduced biomass burning, improvement in the degraded lands, thereby bringing about reduced land erosion. Thus, there could be improved growth in the grass species mixture and these could contribute to GHG mitigation [22].

4.3 The use of decision support tools in beef and dairy cattle management

The measurement of various variables needed in taking decision on cattle performance, health and welfare parameters were carried out by making use of automated devices as outlined by Gonzalez *et al*. and Greenwood *et al*. [23, 24]. Parameters such as cattle liveweight, muscling or body composition could be measured. On-animal sensor devices were also developed to estimate behavioural variables in cattle such as time spent grazing or feeding, ruminating, walking, lying, drinking water and other cattle performance related to health and general animal welfare. Greenwood *et al*. [25] also reported that there were decision support tools for measuring traits in grazing cattle which could be used in genomic and quantitative genetic selection in cattle.

This information could be made available to consumers and policy makers who have influenced in the way that beef and dairy cattle production industries globally were operated for profitable management and to improve productivity, efficiency and sustainability.

4.4 Antibiotic use and health related impacts

The use of antibiotics in the beef and dairy cattle industry as growth promoters needs to be carried out with caution due to anxiety globally over microbial antibiotic resistance [26]. Tona [26] mentioned that the use of antibiotics such as bacitracin, spiramycin and tylosin phosphate as animal feed additives was banned in the European Union (EU) in 1998 and in 2006.

Gott [27] explained that antimicrobial resistance (AMR) occurred when microorganisms, like bacteria were no longer affected by antimicrobial substances (antibiotics) that had previously worked to inhibit their growth or killed them completely.

However, since different organizations world-wide had established rules and regulations to control the use, or to totally ban the application of antimicrobial treatment in livestock in order to reduce the risk of AMR, there were other alternatives suggested as hereby outlined. Cattle should be fed adequate amounts and well balanced feed to support production and to promote good health. The feeding of mycotoxin contaminated feed or feed ingredient, that could hamper the immune function and impair health of the animal should be avoided. It was also suggested that education could be given to livestock farmers or animal health extension agents on the need to practice improved hygiene and good sanitation. The provision of clean water and the observation of farm bio-security practices might also minimize the need for antimicrobial use in livestock. There should also be improved infectious disease control through preventive vaccination. Other researchers [28] stated that in dairy cows, a four layered strategy to reduce antimicrobial use included the following: (i) Appropriate management of animals, farm and soils. (ii) The strategic use of local breeds for cross-breeding with the appropriate exotic breeds. (iii) The conduct of research on the use of herbal and other natural products for the treatment of infected cows. (iv) The feed quality improvement and control.

Beef and dairy cattle health could also be influenced by climatic condition such as temperature related illnesses and deaths [29]. Das *et al*. [30] explained that such temperature related effects could be caused by changes in the immune and endocrine systems. Testa *et al*. [31] also mentioned that seasonal influence on milk somatic cell counts (SCC) could occur in milking cows with increasing SCC values during the summer months. There could also be indirect climatic effects on health due to changes in the intake of concentrate feeds and a decreased forage intake in cattle. These could enhance the development of acidosis which could lead to the development of lameness in cattle. Additionally, the reduction in feed intake in high milk producing cows could increase the risk of experiencing sub clinical ketosis during the summer months [32]. These could be attributed to the fact that high milk yielding cows have high energy requirements for body maintenance and performance that could be frequently mobilized from body nutrients and thus these could lead to lowered level of glucose in the blood and general body weakness in the lactating cows.

4.5 Production cost and returns

McDonald and McBride [1] reported the observation that most of the large dairy enterprises had gross returns that exceeded total cost as compared to mid-sized and small sized farms. These authors [1] further outlined that there were strong incentives for existing large dairies to expand and for producers entering the dairy business to enter in at a large size level. Therefore, it could be deduced that larger farms had substantial cost advantages on the average over smaller operations.

4.6 Governmental policy issues

Some earlier researchers [33] had explained that genomics alone might not bring solutions to genetic improvement needs on the short-term to the developing sectors. However, national strategies such as putting in place adequate livestock extension support services could first of all be required to address the socio-economic issues existing in the various countries.

5. The application of genomics in the practice of beef and dairy cattle production

5.1 Current status of the application of genomics in the practice of dairy cattle production in Ethiopia

Ethiopia was reported to have the largest population (59.5 million heads) of cattle in Africa [34]. This researcher [34] also mentioned in his review article that in Ethiopia, the livestock sector was mainly of smallholder farming system which contributed about 15% of export earnings, 16.5% of the national GDP, 30% of agricultural employment and 35.6% of the agricultural GDP. According to the report of the Central Statistic Agency (CSA) of Ethiopia [35], most of the cattle that were reared by the agrarian community were mostly native breeds which formed about 98.2%; 1.62% of the cattle were crossbreed while the rest 0.18% were of exotic breeds. This author [34] stated that cattle in the country were kept mainly for milk production, draught power, breeding and beef purposes. Most of the milking cows were indigenous animals with low milk yield, 1.37 litres/day, and with an average of 6 months lactation length. Also, the lactating cows had average of age of about 4 ½ years at first calving and about 25 months calving interval. The cattle were mainly grazed on native pasture and given crop residue supplemental feeding. The dairy cattle production systems in Ethiopia included the rural small holder (crop-livestock) dairy production system, the agro pastoral and pastoral dairy cattle production system, the urban and peri-urban smallholder dairy production system and the commercial dairy cattle production system. The commercial dairy farms mostly kept exotic dairy stock. Though Ethiopia was reported [34] to possess the largest livestock (cattle in particular) population in Africa, the animals had low productivity. There had been lack of improvement in the breeding program, uncontrolled mating/breeding practice, feed shortages, prevalence of diseases, and there was generally poor reproductive performance.

There was thus the need to upgrade the reproductive and productive traits in the dairy cattle. Therefore there had been no report of the application of genomics except for the practice of artificial insemination which however, was also faced with the inefficiency of AI technicians and an ineffective animal breeding practice [34].

5.2 Current status of the application of genomics in the practices of beef cattle production in Indonesia

The human population in Indonesia was about 250 million in 2018 while the cattle population was about 16.6m [36]. The country had smallholder cattle production system which formed about 90% of the cattle population and the remaining 10% of cattle were raised by commercial farmers and large beef cattle companies which were usually importing beef from Australia. The large beef cattle companies target market was situated in the Java Island.

Since Indonesia consists of Islands, there was a limited available land space for livestock production and thus there existed a higher demand for cattle meat than the level of production [36]. Agus and Widi [36] pointed out that the government of Indonesia made efforts since the 1980's for the intensive beef cattle production in the country. There had been the promotion of cross-breeding of the local Ongole cattle in Java with high producing *Bos taurus* (Simmental and Limousine) and other European breeds through the use of artificial insemination (AI). However the efforts made through the process of cross-breeding were not successful. In Addition, there had been the issue of unstable government policies on livestock production practices as were also very commonly found to exist in many other developing countries world-wide [36].

Thus there had not been the application of genomics in the breeding practices of beef cattle production in Indonesia. The increasing demand for meat did not match the domestic beef production and the country had no self-sufficiency in meat production [36].

5.3 The application of genomics in South African beef and dairy cattle production sectors to narrow the gap between the developed and developing sectors

In South Africa, the beef and dairy cattle industry had been developed based on the well-developed sector versus the developing sector. The developed sector was made up of the commercial cattle farmers and feedlot companies while the developing sector consisted of the smallholder farmers [33].

Van Marle-Koster and Visser [33] reported that the South African government funded genomic programs were established in the beef and dairy cattle industries in 2015 and 2016. The aim was to set up technological advancement towards moving forward into the application of genomic selection (GS) and a technology driven commercial livestock sector. Blasco and Toro [37] outlined that the phenotyping of some important reproductive and productive traits such as fertility in dairy cows and carcass quality traits in beef cattle for use in genomic selection applications could be of great benefit since these had posed major challenges in the past. The aim of the previous researchers [33] was to bridge the gap in disseminating genetic materials and information to both the farmers in the developed sector and the smallholder farmers in the developing sector in the beef and dairy cattle industry. These could have gone a long way to help lead to an advancement in the application of genomics for sustainable long-term genetic advancement and progress.

South Africa had more than 30 registered beef breeds such as the locally developed Bonsmara composite breed, Nguni, Tuli, Brangus and Simbra breeds [38]. These researchers [38] mentioned that it was only from the locally developed SA Bonsmara breed that the recording of a number of traits such as fertility, growth rate and feed efficiency measuring traits were made. Van Marle-Koster *et al*. [38] stated that for most commonly-measured traits of economic importance, there had been the application of modern selection tools such as the use of estimated breeding value (EBV) by livestock producers. Also under beef production, intensive feedlot testing program results were made available for use in some beef breeds, with data generated for growth rate, feed efficiency and carcass quality measurement traits [38]. In South Africa, the most common exotic dairy cattle breeds were the Holstein and Jersey cattle breeds with average herd sizes of about 400 cow herds and automatic milking system and recording were most commonly used especially in the large dairy herds. Van Marle-Koster and Visser [33] explained that for the emerging and smallholder sectors, genetic tools such as EBVs could not be provided due to small number of animals, incomplete recordings for small number of traits, low percentage recordings and insufficient contemporary groups of animals of similar age groups.

5.4 The application of genomics in beef and dairy cattle breeding in South Africa

Several applications of genomics had become available for beef and dairy cattle farmers [33] in South Africa. Single nucleotide polymorphisms (SNP) arrays of genomics of cattle were widely used in routine genotyping for genomic selection in beef and dairy cattle. These provided added advantage information for using these genotypes for the detection and prediction of carriers of genetic defects [39]. Also the provision of beef and dairy cattle genotypic information could provide the potential for the identification of beneficial genes such as the celtic variant of the polled gene for homozygous polled animals [40].

Some research workers [33] reported that a number of test facilities were available in South Africa for carrying out diagnostic test in ruminants in South African laboratory as shown in **Table 1**. This information however could be useful for the developed commercial industry farmers in particular.

The application of DNA technology could thus serve as a tool for livestock breeders for detecting and removing or culling defective animals from their herds. These could also be used for solving some basic problems while carrying out genetic improvement in the breeding herds [33].

Diagnostic test	Species	South African laboratory
DNA profile	Cattle	Unistel
DNA percentage	Cattle	Unistel
Cytogenetics:1/29 translocation	Cattle	Unistel
Curly calf syndrome	Cattle	Unistel
Polled, horned	Cattle	Unistel
Free-martin	Cattle	Unistel
Unistel, www.unistelmedical.co.za ; source: [33].		

Table 1.
Diagnostic tests available for ruminants in South African laboratory.

The DNA-based percentage verification was however costly and was mainly employed or used in the developed livestock sector. The genomic technology application programs were started in 2015 for beef genomic program (BGP) and in 2016 for the dairy genomic program (DGP) (http://www.livestockgenomics.co.za). These were designed to be funded through the commercial livestock farm industries. Also in the South African livestock sector, DNA marker technology was employed for use under the indigenous farm animal resources conservation [33]. The DNA marker technology was found useful during livestock genetic diversity selection which involved cross-breeding, inbreeding and population structure ecotypes [33].

5.5 Novel traits phenotypes that were used in beef and dairy cattle selection strategies in the smaller developing sectors in South Africa

Genomic selection (GS) was stated to have started in the dairy cattle industry world-wide as phenotypic data and DNA information were made available alongside the use of artificial insemination [41]. The recognition of important or the novel traits associated with sustainability in the dairy industry led to novel traits identification such as feed efficiency (FE), methane emission percentage (%) reduction, heat stress tolerance, claw health, disease resistance and udder health heritability evaluations as shown in the **Table 2** [33].

Novel traits to be used in selection strategies need to be heritable, be of economic importance and should be practically measurable at a cost-efficient level [33].

5.6 Suitable genetic improvement strategies that could be adopted in the developing sector

i. Good quality male and female genetic stock which must be supplied by the seed stock breeders are required. The suitable animals should already be found to contribute to genetic progress [33].

ii. Use of reproductive technology such as AI.

iii. The application of proper recording of individual breeding animal performance.

iv. The formation of farmer co-operative societies where sires such as bulls are shared and AI technicians are employed could be used and disease transmission should be put in check.

Trait	Heritability	Source/References
Feed efficiency		
Residual feed intake (RFI)	0.00–0.40	Egger-Danner *et al.* [42]
	0.01–0.40	Miglor *et al.* [15]
Methane (CH_4) emission reduction	0.09–0.35	Egger-Danner *et al.* [42]
	0.21–0.35	Miglor *et al.* [15]
Claw health		
Hoof lesions	0.02–0.12	Heringstad *et al.* [43]
	0.01–0.13	Miglor *et al.* [15]
Lameness	0.02–0.04	Egger-Danner *et al.* [42]
	0.07–0.15	Heringstad *et al.* [43]
Laminitis	0.06–0.20	Heringstad *et al.* [43]
Disease resistance		
Tick counts	0.03–0.17	Miglor *et al.* [15]
Tick resistance	0.15–0.44	Miglor *et al.* [15]
Heat stress tolerance	0.17–0.33	Miglor *et al.* [15]
Udder health		
Clinical mastitis	0.02–0.09	Egger-Danner *et al.* [42]
Improved somatic cells counts (SCC)	0.01–0.17	Egger-Danner *et al.* [42]

Table 2.
Proposed novel traits for inclusion in selection strategies; source: [33].

v. Care should be taken against indiscriminate crossbreeding between the local and exotic cattle breeds.

vi. Genetic improvement in beef and dairy cattle production in the developing countries need to be backed up by governmental policy strategies which address socio-economic issues such as national livestock extension services support.

vii. In order to reap the benefits of genomics, commercial breeders would need to invest in recording of novel phenotypes and engage in routine genotyping.

6. Future prospects

There is need for effort to be geared globally at bridging the gap between food and nutrition security in the developed and developing beef and dairy cattle production sectors. At the same time, effort could continue to be made towards achieving sustainability goals such as the maintenance of climate change mitigation issues such as GHG emission from the beef and dairy cattle production sectors.

7. Conclusion and recommendations

This review has discussed the semi-intensive and intensive systems of beef and dairy cattle production practiced in the developed countries such as USA, Australia, Europe, Canada, Mexico, China, Japan and South Korea. In the developing countries of Africa and some parts of Asia, however, beef and dairy cattle production was mostly carried out by smallholder farmers under the extensive system.

The low beef and bovine milk production in the developing sectors could be up-graded through controlled crossbreeding of local cattle breeds with the exotic cattle breeds. There should be the use of exotic cattle breeds which have been known to already contribute to genetic progress. These would involve the use of reproductive technologies such as artificial insemination (AI), through farmer co-operatives and where bulls are shared and AI technicians employed.

The application of routine genotyping involving genetic evaluations for most traits of economic importance that could be adopted by breeders in developing countries could be carried out in the breeding laboratory such as found in South Africa. The use of available genetic tools such as estimated breeding values (EBVs), diagnostic tests and DNA percentage testing in selection programs for genetic improvement could be set up in national breeding program laboratories in the developing countries.

More governmental funded programs for genetic development and up-grading of local beef and dairy cattle breeds into more productive animals could be carried out through government extension service support programs. Thus, the continued pursuit of these developmental programs could go a long way in helping to narrow the gap in the developed and undeveloped sectors of the beef and dairy cattle production sectors. Thereby these could lead to the attainment of more intensive, cost effective and sustainable farming systems.

Global agricultural organizations such as the Food and Agricultural Organization (FAO) could continue to make suggestions to the International Livestock Research Institutes world-wide; to keep encouraging research grants to be sought for and monitored to be used for the conduct of local beef and dairy cattle up-grading, through crossbreeding programs in the national agricultural institutions in the developing countries world-wide.

Acknowledgements

The author is grateful to all who helped in one way or another to make this write-up successful.

Author details

Grace Opadoyin Tona
Department of Animal Production and Health, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria

*Address all correspondence to: gotona@lautech.edu.ng

References

[1] MacDonald JM, McBride WD. The Transformation of U.S. Livestock Agriculture Scale, Efficiency and Risks. USDA, Economic Research Service; Dale Simms Editing Support, Washington D.C., USA. 2009. pp. 1-37

[2] Food and Agricultural Organization (FAO). Tackling Climate Change through Livestock: A Global Assessment of Emissions and Mitigation Opportunities. Rome, Italy: FAO; 2013. pp. 1-139

[3] Food and Agricultural Organization (FAO). The Future of Food and Agriculture – Trends and Challenges. Rome, Italy: FAO; 2017. pp. 1-228

[4] Greenwood PL. Review: An overview of beef production from pasture and feedlot globally, as demand for beef and the need for sustainable practices increase. The International Journal of Animal Biosciences: Animal. 2021;**15**:100295

[5] Brito LF, Bedere N, Douhard F, Oliveira HR, Arnal M, Penagaricano F, et al. Review: Genetic selection of high-yielding dairy cattle toward sustainable farming systems in a rapidly changing world. The International Journal of Animal Biosciences: Animal. 2021;**15**:100292

[6] Skelhorn EPG, Gacia A, Nova RJ, Kinston H, Wapenaar W. Public opinion and perception of rose veal in the UK. Meat Science. 2020;**167**:108032

[7] University of Arkansas System (UAS). United States Department of Agriculture, and County Governments Cooperating, Systems of Beef Cattle Production. In: Book-Beef Cattle Production. Arkansas, USA: Division of Agriculture Research and Extension; 2016. pp. 8-10

[8] Greenwood PL, Walmsley BJ, Oddy VH. Regulation of growth and development of skeletal muscle and adipocytes and its impact on efficiency and meat quality. In: Energy and Protein Metabolism and Nutrition. Wagneningen, The Netherlands: Wageningen Academic Publishers. 2019. pp. 53-71

[9] Drouillard JS. Current Situation and future trends for beef production in the United States of America. Asian-Australasian Journal of Animal Sciences. 2018;**31**:1007-1016

[10] Gotoh T, Nishimura K, Kuchida K, Mannen H. The Japanese Wagyu beef industry: Current situation and future prospects-a review. Asian-Australasian Journal of Animal Sciences. 2018;**31**:933-950

[11] Chung KY, Lee SH, Cho SH, Kwon EG, Lee JH. Current situation and future prospect for beef production in South Korea-a review. Asian-Australasian Journal of Animal Sciences. 2018;**31**:951-960

[12] Endres MI, Schwartzkopt-Genswein K. Overview of cattle production systems. In: Tucker CB, editor. Advances in Cattle Welfare. USA: Woodhead Publishing; 2018. pp. 1-25

[13] Rauw UM. Immune response from a resource allocation perspective. Frontiers in Genetics. 2012;**3**:367

[14] Cole JB, VanRaden PM. Symposium review: Possibilities in an age of genomics: The future of selection indices. Journal of Dairy Science. 2018;**101**:3686-3701

[15] Miglior F, Fleming A, Malchiodi F, Brito LF, Martin P, Baes CF. A 100 year Review: Identification and genetic selection of economically important

traits in dairy cattle. Journal of Dairy Science. 2017;**100**:10251-10271

[16] Capper JL, Cady RA, Bauman DE. The environmental impact of dairy production: 1944 compared with 2007. Journal of Animal Science. 2009;**87**:2160-2167

[17] Smith BS, Gotoh T, Greenwood PL. Current situation and future prospects for global beef production: Overview of special issue. Asian-Australasian Journal of Animal Sciences. 2018;**31**(7):927-932

[18] Nieto MI, Barrantes O, Privitello L, Reine R. Green house gas emissions from beef grazing systems in Semi-Arid rangelands of Central Argentina. Sustainability. 2018;**10**:4228

[19] Tona GO. Impact of beef and milk sourced from cattle production on global food security. In: Abubakar M, editor. Bovine Science-Challenges and Advances. London: IntechOpen; 2021. pp.189-204

[20] Herrero M, Thornton PK, Kruska RL, Reid RS. The spatial distribution of methane emissions from African domestic ruminants to 2030. Agriculture, Ecosystems and Environment. 2008;**16**:122-137

[21] Thornton PK, Van de Steeg J, Notenbaert A, Herrero M. The impacts of climate change on livestock and livestock systems in developing countries: A review of what we know and what we need to know. Agricultural Systems. 2009;**101**:113-127

[22] Reid RS, Thornton PK, McCrabb GJ, Kruska RL, Atieno F, Jones PG. Is it possible to mitigate greenhouse gas emissions in pastoral ecosystems of the tropics? Environment, Development and Sustainability. 2004;**6**:91-109

[23] Gonzalez LA, Bishop-Hurley G, Henry D, Charmely E. Wireless sensor networks to study, monitor and manage cattle in grazing systems. Animal Production Science. 2014;**54**:1687-1693

[24] Greenwood PL, Pauli DR, McNally J, Kalinowski T, Ebert D, Little B, et al. Use of sensor-determined behaviours to develop algorithms for pasture intake by individual grazing cattle. Crop and Pasture Science. 2017;**68**:1091-1099

[25] Greenwood PL, Bishop-Hurley GJ, Gonzalez LA, Ingham AB. Development and application of a livestock phenomics platform to enhance productivity and efficiency at pasture. Animal Production Science. 2016;**56**:1299-1311

[26] Tona GO. Current and future improvements in livestock nutrition and feed resources. In: Yucel B, Taskin T, editors. Animal Husbandry and Nutrition. London: IntechOpen; 2018. pp. 147-169

[27] Gott P. Antimicrobial resistance and its practical prevention. Antibiotic reduction-Embracing the challenge. In: All About Feed. Doetinchem, The Netherlands: Misset Uitgeverij B.V. Publisher. 2021. pp.47

[28] Van't Hooft K, Gebru G, Nair MNB, Punniamurthy N, Katushabe E, Groot M. New strategy to reduce antibiotics in dairy farming. Antibiotic reduction-Embracing the challenge. In: All About Feed. Doetinchem, The Netherlands: Misset Uitgeverij B.V. Publisher. 2021. pp. 57

[29] Gauly M, Ammer S. Review: Challenges for dairy cow production systems arising from climate changes. The Animal Consortium. 2020;**14**:s196-s203

[30] Das R, Sailo L, Verma N, Bharti P, Saikia J, Imtiwati I, et al. Impact of heat stress on health and performance of dairy animals: A review. Veterinary World. 2016;**9**:260-268

[31] Testa F, Marano G, Ambrogi F, Borachi P, Casula A, Biganzoli E, et al.

Study of the association of atmospheric temperature and relative humidity with bulk tank milk somatic cell counts in dairy herds using generalized additive mixed models. Research in Veterinary Science. 2017;**114**:511-517

[32] Lacetera N, Bernabucci U, Ranchi B, Nardone A. Body condition score, metabolic status and milk production of early lactating dairy cows exposed to warm environment. Rivista di Agicoltua Subtropicale e Tropicale. 1996;**90**:43-55

[33] Van Marle-Koster E, Visser C. Genetic improvement in South Africa Livestock: Can genomics bridge the gap between the developed and developing sectors? Frontiers in Genetics. 2018;**9**:331

[34] Afras AA. Review on breeding objectives and practices of dairy cattle production in Ethiopia. Journal of Cell and Animal Biology. 2019;**13**(1):1-7

[35] Central Statistical Agency (CSA). Agricultural sample survey, Federal democratic republic of Ethiopia report on livestock and livestock characteristics. (CSA) (2016/17). Available from: http://www.csa.gov.et/survey-report/category/355-eth-agss-2017download=871:report-on-area-and-production-of-major-crops-2010-meher-season

[36] Agus A, Widi TSM. Current situation and future prospects for beef cattle production in Indonesia-A review. Asian-Australasian Journal of Animal Sciences. 2018;**31**(7):976-983

[37] Blasco A, Toro MA. A short critical history of the application of genomics to animal breeding. Livestock Science. 2014;**166**:4-9

[38] Van Marle-Koster E, Visser C, Berry D. A review of genomic selection-Implications for the South African beef and dairy cattle industries. South African Journal for Animal Science. 2013;**43**:1-17

[39] Biscarini F, Schwarzenbacher H, Pausch H, Nicolazzi EI, Pirola Y, Biffani S. Use of SNP genotypes to identify carriers of harmful recessive mutations in cattle populations. BMC Genomics. 2016;**17**:857

[40] Medugorac I, Seichter D, Graf A, Russ I, Blum H, Gopel KH. Bovine polledness-an autosomal dorminant trait with allelic heterogeneity. PLoS One. 2012;**7**:39477

[41] Wiggans GR, VanRaden PM, Cooper TA. The genomic evaluation system in the United States: Past, Present, future. Journal of Dairy Science. 2011;**94**:3202-3211

[42] Egger-Danner C, Cole JB, Pryce JE, Gengler N, Heringstad B, Bradley A. Invited review: Overview of new traits and phenotyping strategies in dairy cattle with a focus on functional traits. Animal. 2015;**9**:191-207

[43] Heringstad B, Egger-Danner C, Charfeddine N, Pryce JE, Stock KF, Kofler J. Invited review: Genetics and claw health: opportunities to enhance claw health by genetic selection. Journal of Dairy Science. 2018;**101**:4801-4821

Chapter 4

Alternative Animal Feeding for Intensive Livestock Farming Systems and Their Impact on Reproductive Performance of Ruminants

Lopes César Mugabe, Maicon Pereira Lents, Emmanuel Emydio Gomes Pinheiro and Rafael Alexandre Muchanga

Abstract

Intensive livestock farming systems are vital for sustaining the growing world population by providing several goods and services. However, the increased livestock operations cost, particularly related to animal feeding, compromises the expansion of this industry, especially in developing countries. One way to reduce the feeding costs without compromising the nutritional quality would be the use of protein-rich food waste discarded by the industries that otherwise would pollute the environment. This chapter presents an overview of the intensive livestock farming systems in developed and developing countries and discusses the use of agro-industrial by-products as alternative sources of nutrients to improve livestock productivity, as well as the key nutritional components that are likely involved to improve the reproductive performance of animals. Our results showed that diets containing 30 to 45% of coconut meal, rich in ether extract and protein, may improve sperm progressive motility, sperm concentration per mL, total sperm per ejaculate, and total viable sperm per ejaculate of beef goats, compared with diets with no or lower coconut meal content. Diets with coconut meals may also enhance the semen quality of sheep.

Keywords: goats, lipid and protein resources, semen quality, sheep, testicular parameters

1. Introduction

Livestock farming is a vital component of agriculture with the potential to promote the economic growth of both developing and developed countries, more so in developing countries. Livestock sustains the world population by providing various services and goods such as meat, milk, eggs, skin, and by-products [1].

Intensive farming refers to a method of food production that relies on intensification techniques. This farming system involves management practices and operations such as concentrated animal feeding, a large number of livestock raised in confinement at high stocking density, and use of selectively bred animals that grow more quickly than naturally occurring breeds and get large enough for slaughter in a shorter period [2]. The intensive livestock farming industry provides employment opportunities for millions of rural households, thereby helping reduce poverty in rural areas, especially in developing countries [2, 3].

By 2050, global livestock production is expected to double-growing faster than any other agricultural subsector, with most of this increase taking place in developing countries [4]. The expansion of the livestock industry may help meet the increasing demand for livestock products, especially in developing countries, which is adding additional pressure on the worldwide livestock farming systems [3]. However, the overall viability of the industrial livestock farming systems depends largely on animal performance and production efficiency, which are negatively affected by the scarcity and fluctuation in the quality and quantity of the animal feed supply. At a certain period of the year, the quality of grazing and browse deteriorates due to seasonal influences, resulting in a decline in animal productivity, unless supplements are provided for the animals [5]. Nonetheless, the conventional feeds commonly used to supplement ruminants are either not available or are available at a high cost, which means that strategies to make livestock farming more efficient to respond to the increasing demand for beef, milk, eggs, and other products are needed. One way to reduce the feeding costs without compromising the nutritional quality would be to use nonconventional feed resources such as by-products from agro-industrial processing and vegetable wastes that are locally available [6]. Many by-products used as alternative feeds are rich in fatty acids such as lauric, myristic, and linoleic, which may enhance productive animal performance [7].

During the dry season in tropical countries, especially in Africa, when only mature senescent material is available, both intake and digestibility are low. As a result, available nutrients do not meet the requirements of the animal, which compromise the animal's ability to reach its genetic potential, unless the nutrient status is increased through supplementation using concentrates that supply primary nutrients such as protein, carbohydrate, and fat [8]. Thus, a better understanding of the quality of by-products and their use in animal feed may probably make livestock farming more efficient, especially in developing countries [9].

This chapter presents an overview of the intensive livestock farming systems in developed and developing countries and discusses the use of agro-industrial by-products as alternative sources of nutrients to improve livestock productivity, as well as the key nutritional components that are likely involved to improve the reproductive efficiency of animals.

2. Global overview of intensive livestock farming systems

Livestock farming throughout the world is becoming increasingly intensive as a result of the increased demand for beef, milk, eggs, and dairy by-products. The estimated population whose livelihood depends on agriculture makes up 42% of the total world population [10]. Therefore, this sector plays a vital role in the process of economic development of any country by ensuring a flow of essential food products, which contribute to food security. Also, livestock farming contributes draught energy and manure for crop production and appears to be the key food and cash security available to many people living in developing countries [4, 11].

It has been reported that the world's population is expected to increase by 2% annually reaching 9.1 billion by 2050, while food demand is projected to increase by 59% in the coming decades. These projections challenge farmers to use proper methods, systems, and technologies to maximize productivity so that they can respond effectively to human consumption needs [12]. Since a majority of rural households live in areas where poverty and deprivation are the most severe and depend on agriculture for their survival, it might seem obvious that livestock needs to be intensive, modern, efficient, and competitive so that its contribution can reduce poverty and transform the economies of the countries [13]. Therefore, efficient and sustainable livestock farming systems in the whole world seem to be a priority for both small and large farmers [14].

Livestock farming worldwide is practiced in three different systems: extensive, semi-intensive, and intensive systems. In extensive livestock farming, mostly practiced in developing countries, the land is individually owned and the animals are heavily dependent on vegetation composed of wild plant species for grazing, which compromises the faster growth of animals and reduces their productive performance [12]. In this system, the animals are only brought indoors and fed meals during lambing and calving seasons or the winter. Although it is a less efficient system, it presents some advantages in terms of sustainability as it returns most of the animal manure directly to the soil, thereby contributing to increased crop production and nutrient cycling [11]. Also, animals can be raised in the semi-intensive systems, where they graze for some period during the day and in the evening, they feed on supplements. Both extensive and semi-intensive systems are practiced by smallholder farmers, and they are more suitable for ruminants such as cattle, goats, and sheep [15].

Currently, in the whole world, more so in developed countries, farmers are practicing intensive system-based management so that they can provide beef, milk, and eggs throughout the year. In this system, also described as factory farming, animals are enclosed in zero-grazing units where are fed and provided water to yield high productivity (beef, dairy produce, and poultry) from limited land available by raising large numbers of animals indoors. Under this system, diet, breeding, and disease are managed precisely and in consequence, meat, milk, and other products are produced in large quantities [16]. However, confined animals are usually fed based on concentrates composed of very expensive feedstuffs which makes this system the most expensive, requiring high initial costs to develop the activity [17]. One of the key strategies to make intensive livestock farming cheaper would be the adoption of food alternatives based on locally available and low-cost by-products such as coconut meal, palm kernel cake (PKC), palm fiber, cottonseed meal, and other vegetable and fruit wastes [18].

3. Alternative animal feeding for intensive livestock farming

One of the major constraints to livestock farming is the scarcity and fluctuation of the quantity and quality of the feed supply. However, it has been estimated that about one-third of the food produced globally for human consumption is wasted, representing a significant loss of the resources spent making and processing food and a threat to food security. The reduction of food waste is, therefore, a potential strategy for closing the gap between the supply and demand for food [19]. Moreover, competition between humans and animals for grains such as corn, wheat, bean, soybean, and others is increasing. Thus, it has become imperative to consider the use of food waste rich in protein, lipids, and some essential nutrients for animal health as feedstock in the animal feed industry [20].

Food waste is also referred to as a nonconventional feed resource, which is defined as feeds that have not been traditionally used in animal feeding and/or are not normally used in commercially produced rations for livestock. Some of such nonconventional feeds are agro-industrial by-products, which contain little economical value as foods for human consumption. In recent years, agro-industrial by-products have become major sources of dietary nutrients and energy in support of beef and milk production because they are available for use as livestock feeds at competitive prices relative to other commodities [21, 22]. Thus, the use of food waste for livestock feeding can help farmers reduce feed costs and help food waste generators reduce disposal costs while minimizing the environmental impacts of this waste [20].

In developing countries, the productivity of ruminant livestock has been constrained by the low quality of feed supply, since the basal diet of most cattle, sheep, and goats almost totally consists of roughage. Farmers often use high amounts of cereal grain-based concentrates for meat and milk production; however, this practice has introduced competition since grain is used for human consumption [23, 24]. Thus, providing adequate good quality feed to livestock to raise and maintain their productivity may be a major challenge to farmers all over the world. Future hopes of feeding millions of people worldwide and safeguarding their food security will directly depend on a high level of productivity, animal performance, and efficiency of the livestock farming, which may be reached through intensive animal farming that includes supplementary feeding plans based on by-products feeds [1].

In some developing countries in Africa, the farmers can neither spare land for feed production nor can they afford to buy expensive concentrates to feed animals. Therefore, efficient utilization of nonconventional feed for ruminants is a priority. For goats and sheep, supplementary feeding could be performed using crop residues, hay or silage for energy, protein concentrates, and agro-industrial by-products for additional protein [18]. So, it is important to support farmers with knowledge and technologies related to alternative feed resources that are easy to adopt and economically viable. To that end, studies were performed in Brazil to assess the nutritional composition of coconut meals and their impact on the reproductive performance of goats under the grazing system, and the results are discussed in the next section.

3.1 Nutritional composition of by-products used as alternative feeding for ruminants

The limited supply of nutritious feed is one of the main limiting factors to efficient animal production and reproduction. An intensive feeding system based on locally available by-product feeds may be an alternative promising feeding system to rear ruminants economically. However, the chemical composition of agro-industrial by-products may vary largely depending on the feedstock type (**Table 1**).

Some nonconventional feeds such as coconut meal contain a crude protein ranging from 20 to 25% dry matter (DM) with relatively high quantities of cell wall constituents [neutral detergent fiber (NDF) more than 50% DM, and acid detergent fiber (ADF) of about 30% DM] [49]. Coconut meal obtained from mechanical extraction has generally high oil content (about 5 to 15% DM). This quantity of oil content makes it a valuable energy source for ruminants. An increase in the energy content of the

Agro-industrial by-products	Nutritional composition[z] (%)							Reference
	DM	Ash	CP	EE	ADF	CF	NDF	
Palm fiber	97.60	7.50	7.40	7.70	40.0	37.50	66.80	Padilla et al. [25]
	91.00	5.30	ND	2.69	54.80	ND	76.80	Obese et al. [26]
	95.30	5.30	15.12	3.98	25.30	ND	46.30	Chanjula et al. [27]
Chickpea	89.60	6.80	6.50	1.20	51.60	39.00	69.40	Bampidis et al. [28]
	89.30	3.30	21.90	4.00	4.80	ND	27.70	Serrapica et al. [29]
	91.10	2.85	22.50	5.09	ND	1.64	ND	Majewska et al. [30]
Soybean meal	93.85	5.70	44.56	5.69	7.35	5.60	13.84	NRC [31]
	88.87	6.39	51.41	3.45	10.13	6.18	12.22	Zambom et al. [32]
	87.9	7.21	54.30	1.32	7.00	ND	9.10	Harper et al. [33]
Orange pulp	22.10	ND	15.10	19.10	17.20	7.70	26.50	Minguez et al. [34]
	90.00	6.00	11.00	12.00	14.00	7.86	41.00	Watanabe [35]
Rapeseed meal	93.11	6.39	35.19	9.97	17.57	9.77	23.77	NRC [31]
	88.0	7.30	38.60	1.40	16.80	11.80	20.70	Feng and Zuo [36]
	88.70	ND	33.70	2.30	19.60	12.40	28.30	Maison [37]
Palm kernel cake	95.8	3.06	10.05	8.21	13.48	19.30	71.37	Mugabe et al. [38]; Suhaimi et al. [39]
	91.87	3.53	13.97	10.78	56.02	ND	64.09	Bringel et al. [40]
	95.29	3.33	16.64	7.78	45.71	ND	70.04	Oliveira et al. [41]
Cottonseed meal	90.69	6.39	39.22	5.50	17.92	13.96	25.15	NRC [31]
	94.50	7.51	24.28	7.94	31.80	ND	43.56	Tavares-Samay et al. [42]
	92.00	5.00	23.00	17.50	20.00	20.80	40.00	Blasi and Drouillard [43]
Apple pomace	92.35	ND	6.62	5.53	20.78	14.48	39.03	Xiong et al. [44]
	95.20	2.30	9.02	3.70	46.70	ND	61.20	Mirzaei-Aghsaghali et al. [45]
	88.84	6.10	7.70	6.18	43.20	51.10	68.50	Beigh et al. [46]
Coffee pulp	94.30	5.80	12.90	2.40	ND	14.60	37.20	Padilla et al. [25]
	69.16	16.6	18.12	5.71	ND	33.63	ND	Daniel et al. [47]
	88.69	8.42	12.58	2.65	77.84	ND	54.39	Barcelos et al. [48]

[z]*DM, dry matter; CP, crude protein; EE, ether extract; NFE, nitrogen-free extract; CF, crude fiber; ADF, acid detergent fiber; NDF, neutral detergent fiber. ND, not determined. The values are presented as a percentage of the dry mater content.*

Table 1.
Chemical composition of different by-products from agro-industrial processing.

diets through supplementary concentrate can potentially enhance milk production and improve growth rate, body weight gain, and reproductive performance [50, 51].

There are further feedstuffs also valuable as an alternative feed for ruminants, such as palm fiber, chickpea, orange pulp, rapeseed meal, cottonseed meal, and palm kernel cake (PKC), which because of its nutritional content may be described as an energy food. In addition, palm kernel cake supplies protein and energy but is usually classified as a source of protein, which may vary from 10.1 to 16.6% (**Table 1**). This protein content of PKC is regarded as sufficient to meet the needs of most ruminants [52, 53]. Proteins are the principal constituents of the organs and muscles and are required by ruminants for metabolic functions [38, 54]. Palm kernel cake also contains a large amount of crude fiber (CF; 19.3%), dry matter ranging from 91.9 to 95.8%, ether extract (EE; 7.8 to 10.8%), ash (3.1 to 3.5%), neutral detergent fiber (NDF; 64.1 to 71.4%), acid detergent fiber (13.5 to 56.0%) (**Table 1**), nitrogen-free extract (NFE; 46.7 to 58.8%), total carbohydrates (78.7%), nonfibrous carbohydrates (NFCs) (7.31%), hemicellulose (HEM) (25.98%), and lignin (4.28%) [38, 55]. The metabolizable energy (ME) of palm kernel cake for ruminants is 2.5 to 2.6 Mcal kg^{-1}, which is considered suitable for most ruminants [55].

Animals must be fed diets containing the earlier-mentioned essential nutrients for healthy growth and increased production. The ether extract is the main source of lipids which constitutes the principal component of sperm cells. It also stimulates cholesterol biosynthesis and testosterone secretion to sustain semen production in males [56]. Other fibrous components are sources of carbohydrates which are converted to acetic acids and used by the cows for energy and precursor of fat in milk [57]. It is also essential for animal health, since it is required by ruminants to support an appropriate rumen function and physiology [58].

3.2 Dietary effects on reproductive performance of ruminant livestock

The process of reproduction is aligned closely with the food supply. It is a coordinated function of many tissues, cell types, and regulatory systems which is possible only when animals are provided with sufficient quantities of dietary nutrients [59]. In tropical regions such as Africa, ruminants raised on smallholder farms face constraints of improper nutritional management, which may negatively affect their growth, health, and reproductive performance [60, 61]. For instance, puberty is usually delayed in grazing ruminants because grasslands in semiarid zones in the tropics do not provide enough protein and other nutrients, which leads to insufficient production of rumen microbial protein to support optimum growth rate [60]. Thus, nutrition plays a pivotal role in maintaining the body condition and reproductive efficiency of animals.

Nutrition consists of different nutrients such as protein, fat, carbohydrates, and micro-elements. Carbohydrates and proteins provide substrates for rumen fermentation, which results in the production of volatile fatty acids (VFAs). Ruminants utilize these VFAs as their main source of energy for maintenance, milk production, and reproductive performance. Malnutrition resulting from inadequate, excess, or imbalanced nutrient intake may lead to the loss of body weight. Low body condition delays the onset of puberty (up to 1 year in sheep and goats), reduces ovulation and lower conception rates, compromises embryonic and fetal survival, increases the postpartum interval to conception, interferes with normal ovarian cyclicity by decreasing gonadotropin secretion, increases the chances of infertility, and influences the rate of genetic progress [62, 63]. So, it seems to be clear that the initial reproductive events are sensitive to fluctuations in nutrient availability [61].

An inverse relationship between growth rate and age at puberty also exists in males. Low-quality feeding has been shown to delay puberty by about 5 months in bulls. The animals had poor testicular development and smaller ejaculates as compared to their normal counterparts. On the other hand, bulls kept on grazing plus concentrate feed showed a better growth rate than those on grazing only, with an earlier age at puberty and a greater scrotal circumference [60]. Even at maturity, the size of the testicles of underfed animals can be smaller [59] resulting in a lower sperm concentration in the semen. It has also been reported that the seminal vesicles contain less fructose and citric acid as well as smaller Leydig cells and seminiferous tubules and decreased testosterone levels under protein stress conditions [64]. Restriction of both protein and energy also prevented Merino lambs from reaching puberty in their first potential breeding season, with reduced testicular size and sperm quality in the ejaculate. Feed restriction also reduces libido in rams [60].

3.2.1 Effect of coconut meal on goat reproductive performance

To assess the effectiveness of coconut meal as an alternative feed and its effect on beef goats' reproductive performance, a study was conducted in Brazil in 2016. Forty-eight males with initial body weight ranging from 26.04 to 28.54 kg were assigned in a completely randomized design into four groups with 12 replicates each under a semi-intensive system. The animals were kept indoors during the night and moved to graze in the morning on a pasture composed of *Brachiaria decumbens (*nonlegume forage plant). In the afternoon, the animals were supplemented with a concentrate containing 0% of coconut meal (group 1), 15% of coconut meal (group 2), 30% of coconut meal (group 3), and 45% of coconut meal (group 4) (**Table 2**). Before feeding the animals, the nutritional composition of coconut meal, soybean bagasse, corn meal, and pasture was assessed for dry matter, ash, ether extract, crude protein, neutral detergent fiber, total carbohydrates, hemicellulose, and lignin, and the results are shown in **Table 3**. In our study, coconut meal was included in four different levels (0%, 15%, 30%, and 45% of coconut meal) as a source of fatty acids and protein as well as a substitute for soybean meal, which is one of the most expensive feedstuffs in animal feed. In addition, the levels of cornmeal were also reduced while increasing the level of coconut meal. The animals' body weight and testicular parameters were recorded every 15 days, and the semen was collected and assessed for volume, wave motion, progressive motility, sperm concentration, and sperm vigor according to the methodology suggested by CBRA [65]. In addition, the total number of sperm and the total number of viable spermatozoa were estimated according to the methodology described by Martin-Rillo [66].

Our results showed an increase in the scrotal circumference and testicular volume of goats fed with concentrates containing 30% and 45% of coconut meal (**Table 4**). This result suggests that concentrates with high amounts of coconut meal may be more effective than those with lower amounts in improving goat's testis. Also, the data in **Table 4** suggest that scrotal circumference and testicular volume may increase as the body weight of goat increases, indicating a positive relationship between nutrition and goats' testis size or reproductive efficiency. Males with a larger testis tend to produce more semen and offspring that reach puberty at an earlier age and release more ovules during each estrous period [68]. Oldham et al. [69] found that a 25% increase in testicular size led to an 81% increase in the production of spermatozoa. Also, Cameron et al. [70] found that an 86% increase in testicular size led to a 250% increase in the production of spermatozoa. The positive relation between testicular

Ingredients	Level of coconut meal (%)			
	0	15	30	45
		g kg^{-1} as fed		
Coconut meal	0	150	300	450
Soybean meal	350	320	290	250
Cornmeal	600	450	350	250
Mineral salt	20	20	20	20
Urea	30	30	30	30

Table 2.
Level of coconut meal (treatments) in diet supplied to the goats.

Nutrients	Ingredients			*Brachiaria decumbens*
	Coconut meal	Soybean bagasse	Corn meal	
		%		
Dry matter	90.45	87.71	86.45	41.60
Ash	3.76	7.05	3.76	6.31
Ether extract	13.79	3.11	4.95	2.38
Neutral detergent fiber	58.46	57.60	30.69	52.01
Crude protein	23.57	46.38	6.74	7.48
Total carbohydrates	4.89	42.95	86.40	76.13
Hemicellulose	24.95	49.31	28.71	18.55
Lignin	16.21	2.96	1.47	3.09

Source: Mugabe et al. (unpublished).

Table 3.
Chemical composition of forage and feedstuffs used in concentrated feed for goats in 2016.

biometric parameters and body weight concerning nutrition effects was also reported by Martinez et al. [71].

The diets with 30% and 45% of coconut meal showed higher levels of ether extract (**Table 5**) which is a saturated fatty acids source, such as lauric (C12:0), myristic (C14:0), palmitic (C16:0), and stearic (C18:0) acids and unsaturated fatty acids such as myristoleic (C14:1), palmitoleic (16,1), oleic (C18:1), and polyunsaturated fatty acids (PUFA) that include linoleic (C18:2) and linolenic (C18:3) acids, compared with other diets. So, it may be possible that the fatty acids in coconut meal may have increased the concentration of serum cholesterol which may lead to improved testosterone levels in goats. However, changing nutrition in small ruminants alters not only the total mass of testicular tissue but also the efficiency with which the spermatozoa are produced by that tissue [77].

It has been classically admitted that in both the mid and the long terms, variations in growth, testicle parameters, and fluctuations in live weight as a result of nutritional status affect also semen quality of ruminants, as nutrition appears to be the major modulator of sexual efficiency in small ruminants [71]. In our previous study with sheep managed under extensive range grazing conditions and fed with palm kernel cake as an

Parameters	Levels of coconut meal (%)			
	0	15	30	45
Initial body weight (kg)	27.38 ± 0.49	26.04 ± 57	27.50 ± 1.20	28.54 ± 2.41
Final body weight (kg)	35.10 ± 2.4[b]	37.01 ± 1.1[b]	45.02 ± 1.3[a]	47.08 ± 2.6[a]
DM intakeZ	113.49 ± 4[b]	127.03 ± 4[b]	159.10 ± 3[a]	157.39 ± 8[a]
Body score	3.20 ± 0.45	3.15 ± 0.20	3.70 ± 0.95	3.85 ± 0.65
Scrotal circumferenceY (cm)	25.29 ± 2.8[b]	24.33 ± 3.9[b]	36.40 ± 1.7[a]	38.05 ± 1.2[a]
Length of right testis (cm)	7.43 ± 1.39	7.81 ± 1.80	8.40 ± 1.53	8.11 ± 1.04
Width of right testis (cm)	4.89 ± 0.33	4.68 ± 0.81	5.20 ± 0.92	5.48 ± 0.99
Length of left testis (cm)	7.11 ± 1.43	7.90 ± 1.56	8.31 ± 1.00	8.05 ± 1.29
Width of left testis (cm)	4.19 ± 0.02	4.50 ± 0.63	5.10 ± 0.7	5.30 ± 0.42
Testicular volumeX (cm)	223 ± 109[b]	249 ± 128[b]	310 ± 104[a]	342 ± 101[a]
Testis shapeV	0.55 ± 0.0	0.58 ± 0.0	0.60 ± 0.0	0.59±0.0

[Z]*Dry matter intake (g/day).*
[Y]*Scrotal circumference was recorded using a measuring tape around the widest point of the testicles.*
[X]*Testicular volume (cm^3) was estimated using the formula for a cylinder, where $V=2\times r^2 \times \Pi \times h$; r = testis width/2; Π (Pi) = 3.14; h, testis length.*
[V]*The testis shape was recorded by the ratio between the width and length of the right and left testicles according to the methodology described by [67], which classifies testicles from long (ratio 0.5), long moderate (ratio 0.51 up to 0.625), long oval (ratio 0.626 up to 0.750), oval-spherical (ratio from 0.751 up to 0.875) to spherical shape (ratio equal or greater than 0.876).*
Means followed by the same letters in each row are not significantly different at 5% by the Tukey test.
Source: Mugabe et al. (unpublished)

Table 4.
Testicular parameters and initial and final body weight of goats fed coconut meal in 2016.

NutrientsZ	Levels of coconut meal (%)			
	0	15	30	45
		$g\ kg^{-1}$		
Dry matter	710.38	718.40	739.02	741.38
Ether extract	42.50	43.28	51.39	56.19
Mineral matter	34.20	36.47	37.18	37.04
Crude protein	212.40	195.03	200.41	190.47
Neutral detergent fiber	240.60	320.31	345.02	380.58
Non-fiber carbohydrates	482.49	436.07	395.72	329.36
Total digestible nutrients	825.40	810.42	790.05	722.48
Cellulose	22.39	26.10	68.55	94.05
Hemicellulose	320.31	354.04	349.58	337.04
Lignin	52.30	67.54	86.22	92.30

[Z]*Chemical analysis for dry matter (DM), mineral matter (MM), crude protein (CP), and ether extract (EE) was performed according to the procedure suggested by AOAC [72]. Neutral detergent fiber (NDF) was determined using the method recommended by Van Soest et al. [73]. Non-fiber carbohydrates (NFCs) were estimated using the following equation, as suggested by Hall [74]: NFC = 1000-(NDF+CP+EE+MM). Hemicellulose (HEM) and cellulose (CEL) were obtained by the sequential method, according to the methodologies recommended by Silva and Queiroz [75] using the following equations: HEM=NDF-ADF and CELL=ADF-Lig. (lignin), respectively. Total digestible nutrients (TDNs) were determined using an equation according to NRC [76]: TDN=87.84-(ADF × 0.70).*
Source: Mugabe (unpublished)

Table 5.
Nutritional composition of the concentrates fed to goats in 2016.

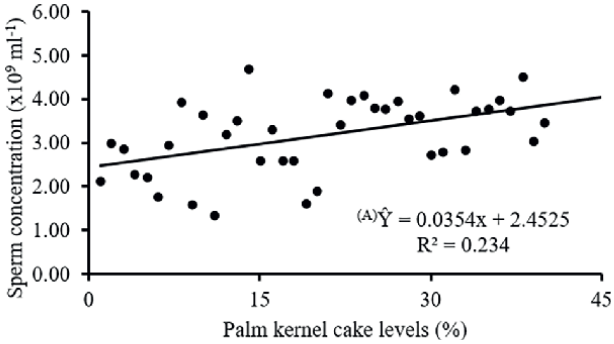

Figure 1.
Sperm concentration (×10^9 mL^{-1}) of lambs fed diets containing levels of palm kernel cake. Source: Mugabe et al. [38].

Seminal parameters	Level of coconut meal (%)			
	0	15	30	45
Volume (mL)	1.92 ± 0.45	1.87 ± 0.59	1.90 ± 0.3	1.96 ± 0.36
Mass motility (0–5)	3.00 ± 2.0	3.50 ± 1.0	3.00 ± 1.35	3.00 ± 1.00
Progressive motility (%)	60.4 ± 3.0^b	65.0 ± 4.0^b	80.5 ± 2.0^a	85.0 ± 0.0^a
Spermatic vigor (0–5)	3.00 ± 2.0	3.00 ± 1.00	3.00 ± 2.00	3.00 ± 0.00
Concentration (×10^9 mL^{-1})	1.45 ± 1.0^b	1.75 ± 1.82^b	3.91 ± 2.47^a	3.55 ± 1.90^a
Total sperm/ejac. (×10^9)^z	1.39 ± 1.0^b	1.55 ± 1.93^b	2.90 ± 1.44^a	2.87 ± 1.47^a
Viable spermat./ejac. (10^9)^z	1.15 ± 0.5^b	1.20 ± 0.08^b	1.83 ± 0.37^a	1.95 ± 0.21^a

^{Z}*ejac., ejaculate.*
Means followed by the same letters in each row are not significantly different at 5% by the Tukey test.
Source: Mugabe (unpublished)

Table 6.
Physical parameters of the semen of goats fed with coconut meal under grazing in 2016.

alternative fatty acids source, it was observed that the diets affected sperm concentration of animals, more so diets containing 45% of palm kernel cake (**Figure 1**). Our results suggested positive relation between sperm concentration and nutrition, which means that animal feed supplementation is a key strategy to raise the reproductive performance of ruminants, as larger scrotal circumference, greater ejaculate volume, higher sperm concentration, higher daily sperm production as well as better seminal quality are strongly associated with nutritional management [38].

In the current study, diets containing 30% and 45% of coconut meal improved sperm concentration per mL, total sperm number per ejaculate as well as sperm progressive motility when compared to the non-supplemented group (**Table 6**). Our results agreed with those of Santos et al. [78] who found that palm kernel cake and coconut meal affected positively the sperm concentration. Moreover, Rege et al. [79] found a strong influence of diets containing fatty acids source on sperm concentration. Depending on the age, genetics, and nutritional status, sperm concentration may vary from 1 to 3 × 10^9 spermatozoa mL^{-1} [80]. These results are similar to those observed in our study in which the sperm concentration ranged from 1.45 × 10^9 to 3.55 × 10^9 spermatozoa mL^{-1}.

It has become apparent that reproductive efficiency depends on an adequate nutritional plan. However, other physical parameters of the semen of goats such as semen volume, mass motility, and spermatic vigor did not vary significantly with diets. Nonetheless, the findings from other researchers [81, 82] showed greater semen volume from lambs supplemented with a concentrate containing wheat bran, crushed maize, soybean meal, fish meal, salt, and vitamin-mineral premix than the control. These opposed results may possibly be related to the differences in the crude protein content of the concentrates used in the studies.

4. Conclusion

Intensive livestock farming may be a great strategy to ensure global food security and meet the increased global demand for meat, eggs, and dairy products. However, high operations cost, particularly related to animal feeding, compromises the expansion of this vital industry, especially in developing countries. The use of agro-industrial by-products may help reduce feeding costs without compromising the nutritional quality while reducing the risk of environmental pollution. However, by-products feeds vary largely in nutritional composition, with some containing high levels of crude protein, ether extract, and carbohydrates which have a significant influence on the growth and reproductive performance of ruminants. Diets containing 30 to 45% of coconut meal, rich in lipids and protein, may improve sperm progressive motility, sperm concentration per mL, total sperm per ejaculate, and total viable sperm per ejaculate of beef goats, compared with diets with no or lower coconut meal content. Diets with coconut meals may also enhance the semen quality of sheep. Thus, the use of coconut meal for animal feeding can make intensive farming systems more efficient by improving animal production and reducing feeding costs.

Acknowledgements

We acknowledge the financial support provided by the National Research Fund (Fundo Nacional de Investigação - FNI), and the Ministry of Science, Technology, and Higher Education of Mozambique through the Scientific Initiation Scholarships.

Conflict of interest

"The authors declare no conflict of interest."

Author details

Lopes César Mugabe[1], Maicon Pereira Lents[2], Emmanuel Emydio Gomes Pinheiro[2] and Rafael Alexandre Muchanga[1*]

1 Faculty of Agronomic Engineering and Forestry, Zambeze University, Mocuba, Mozambique

2 Center for Agrarian, Environmental and Biological Science, Federal University of Recôncavo of Bahia, Brasil

*Address all correspondence to: rafael.muchanga@uzambeze.ac.mz

References

[1] Makkar H. Animal Production and Health Division; 2005

[2] Upton M. The role of livestock in economic development and poverty reduction. Research Gate; 2004

[3] Herrero M, Thornton PK. Livestock and global change: Emerging issues for sustainable food systems. Proceedings of the National Academy of Sciences. 2013;**110**:20878-20881

[4] Ilea R. Intensive livestock farming: Global trends, increased environmental concerns, and ethical solutions. Journal of Agriculture and Environment Ethics. 2009;**22**:153-167

[5] Makkar HPS. Improving Animal Production by Supplementary Feeding of Multinutrient Blocks, Controlling Internal Parasites and Enhancing Utilization of Alternate Feed Resources. Vienna, Austria: IAEI; 2006

[6] Moyo SB, Swanepoel FJC. Multifunctionality of Livestock in Developing Communities. In: Swanepoel F, Stroebel A, Moyo S, editors. The role of livestock in developing communities: Enhancing multifunctionality. University of the Free State and Technical Centre for Agricultural and Rural Cooperation (CTA), Cape Town, South Africa. 2010. pp. 1-12

[7] Gowe C. Review on potential use of fruit and vegetables by-products as a valuable source of natural food additives. Food Science and Quality Management. 2015;**45**:47-61

[8] Smith OB, Somade B. Nutrition reproduction interactions in farm animals. In: Paper presented at the International Foundation seminar on Animal Reproduction. Niamey Niger; 1994

[9] Potts GR. Application of research results on by-product utilization: Economic aspects to be considered. In: Proceedings of a Workshop on Applied Research held in Nairobi, Kenya. 1982

[10] FAO (Food and Agriculture Organization). The Future of Food and Agriculture – Trends and Challenges. Rome: FAO; 2017

[11] Jahnke HE. Livestock Production System and Livestock Development in Tropical Africa. Kiel: KWV; 1982

[12] Moekti GR. Industrial livestock production: A review on advantages and disadvantages. IOP Conference Series: Earth Environmental Science. 2020:492

[13] Diao X, Hazell P, Trurlow J. The Role of Agriculture in African Development. World Development. 2010;**38**:1375-1383

[14] Pawlak K, Kolodziejczak M. The role of agriculture in ensuring food security in developing countries: Considerations in the context of the problem of sustainable food production. Sustainability. 2020;**12**:5488. DOI: 10.3390/su12135488

[15] Mubiru R, Namirimu T, Owino S, Kyalingonza L, Nyadoi P, Buyinza J. From Extensive to Semi-intensive Livestock Production Systems in Ugand's Albertine Rift: Practical Interventions Manual. Uganda Wildlife Society and Buliisa Distrit Local Government, Uganda; 2013

[16] Kasapidou E, Basdagianni Z, Papadopoulos V, Karaiskou C, Kesidis A, Tsiotsias A. Effects of intensive and semi-intensive production on sheep milk chemical composition, physicochemical characteristics, fatty acid profile, and nutritional indices. Animals. 2021;**11**:2578

[17] Manousidis T, Abas Z, Ragkos A, Abraham EM, Parissi ZM, Kyriazopoulos AP. Effects of the economic crisis on sheep farming system: A case study from the north Evros region, Greece. In: Acar Z, López-Francos A, Porqueddu C, editors. New Approaches for Grassland Research in Context of Climate and Socio-economic Changes. Zaragoza: CIHEAM; 2012. pp. 439-442

[18] Degang Z, Xiaopeng PXL, Xiaojing L. Evaluation of tree forages as non-conventional feeds in gansu province of China; 2006.

[19] FAO. Moving Forward on Food Loss and Waste Reduction. Rome, Italy: FAO; 2011

[20] Rivin J, Miller Z, Matel O. Using Food Waste as Livestock Feed. University of Wisconsin-Extension, Madison, USA; 2014

[21] Devendra C. Non-conventional Feed Resources in Asia and Pacific. 2nd ed. Food and Agriculture Organization of the United Nations, APHCA, Bangkok; 1985

[22] Mirzaei-Aghsaghali A, Maheri-Sis N. Nutritive value of some agro-industrial by-products for ruminants – a review. World Journal of Zoology. 2008;**3**:40-46

[23] Yang K, Qing Y, Yu Q, Tang X, Chen G, Fang R, et al. By-products feeds: Current understanding and future perspectives. Agriculture. 2021;**11**:207

[24] Akbar MA, Ahmed TU, Mondal MMH. Improving animal productivity and reproductive efficiency: Development and testing medicated urea-molasses multi-nutrient blocks in rural farms of Bangladesh; 2006.

[25] Padilla DJG, La Plata RD, Curi LMF, Mendiola AEL, Alcarraz RER, Lozano VH, et al. Characterization of the nutritional value of agro-industrial by-products for cattle feeding in the San Martin region. Science and Agriculture. 2020;**21**:1374

[26] Obese FY, Osafo ELK, Okai DB. Evaluation of the feeding value of palm press fibre using in vitro digestibility techniques. Tropical Animal Health and Production. 2001;**33**:165-172

[27] Chanjula P, Petcharat V, Cherdthong A. Rumen characteristics and feed utilization in goats fed with biologically treated oil palm fronds as roughage in a total mixed ration. South African Journal of Animal Science. 2018;**48**:1049-1056

[28] Bampidis VA, Christodoulou V. Chickpeas (*Cicer arietinum L.*) in animal nutrition: A review. Animal Feed Science and Technology. 2011;**168**:1-20

[29] Serrapica F, Masucci F, Romano R, Napolitano F, Sabia E, Aiello A, et al. Effects of chickpea in substitution of soybean meal on milk production, blood profile and reproductive response of primiparous buffaloes in early lactation. Animals. 2020;**10**:515

[30] Majewska AD, Kwiecien M, Mieczan AW, Haliniarz M, Bielak A. Raw chickpea (*Cicer arietinum* L.) as a substitute of soybean meal in compound feed for bloiler chickens: Effects on growth performance, lipid metabolism, fatty acid profile, antioxidant status, and dietary value of muscles. Animals. 2021;**11**:3367

[31] National Research Council (NRC). Critical Role of Animal Science Research in Food Security and Sustainability. Washington, DC, USA: The National Academies Press; 2012

[32] Zambom MA, dos GT S, Modesto EC, Alcalde CR, Gonçalves GD, da DC S, et al.

Nutritional value of soybean hulls, soybean meal, ground corn and wheat meal for cattle. Acta Scientiarum. 2001;**23**:937-943

[33] Harper MT, Melgar JO, Nedelkov K, Raisanen S, Chen X, Martins CMMR, et al. Production effects of feeding extruded soybean meal to early-lactation dairy cows. American Dairy Science Association. 2019;**102**:8999-9016

[34] Minguez C, Calvo A. Effect of supplementation with fresh orange pulp (*Citrus sinensis*) on mortality, growth performance, slaughter traits and sensory characteristics in meat guinea pigs. Meat Science. 2018;**145**:51-54

[35] Watanabe PH, Thomas MC, Ruiz US, Santos VM, Masson GC, Fraga AL, et al. Carcass characteristics and meat quality of heavy swine fed different citrus pulp levels. Arquivo Brasileiro de Medicina Veterinária e Zootecnia. 2019;**62**:921-929

[36] Feng D, Zuo J. Nutritional and anti-nutritional composition of rapeseed meal and its utilization as a feed ingredient for animal. Feed and Industrial raw material: Feed. 2003;**37**:265-270

[37] Maison TW. Evaluation of the nutritional value of canola meal, 00-rapeseed meal, and 00-rapeseed expellers fed to pigs. Dissertation, University of Illinois; 2013.

[38] Mugabe LC, Bagaldo AR, Barbosa LP, Araújo FL, Oliveira BYS, Silva RVMM, et al. Biochemical and seminal parameters of lambs fed palm kernel cake under grazing system. Revista Brasileria de Zootecnia. 2017;**46**:670-677

[39] Suhaimi D, SMA S. Nutritional value variation in local palm kernel cake for animal feeds. Malaysian Journal of Veterinary Research. 2018;**9**:165-170

[40] Bringel LML, Neiva JNM, Araújo VL, Bomfim MAD, Restle J, Ferreira ACH, et al. Consumo, digestibilidade e balanço de nitrogênio em borregos alimentados com torta de dendê em substituição à silagem de capim-elefante. Revista Brasileira de Zootecnia. 2012;**40**:1975-1983

[41] Oliveira RL, Ribeiro OL, Bagaldo AR, Lima LS dos, Borja MS, Correia BR, Costa JB, Leão AG. Palm kernel cake from biodiesel production on Massai grass ensilage. Revista Brasileira de Saúde Producao Animal. 2011;**12**:881-892

[42] Tavares-Samay AMA, Junior WMD, Palhares LO, Lopes CC, Rabello CBV, AHSC C. Determination of nutrient and energy values of cottonseed meal supplemented or not with phytase and protease for broiler chicks. Brazilian Journal of Animal Science. 2019;**48**:e20180142

[43] Blasi DA, Drouillard J. Composition and Feeding value of cottonseed feed products for beef cattle. K-States. 2002;**2**:426

[44] Xiong ML, Wu XJ, Zhu XF, Zhang WJ. Effects of apple pomace on growth performance, organ indexes and serum biochemical indexes of guanzhong dairy goats. China Journal of Animal Nutrition. 2020;**32**:2683-2689

[45] Mirzaei-Aghsaghali A, Maheri-Sis N, Mansouri H, Razeghi ME, Shayegh J, Aghajanzadeh-Golshani A. Evaluating nutritional value of apple pomace for ruminants using in vitro gas production technique. Annals of Biological Research. 2011;**2**:100-106

[46] Beigh YA, Ganai AM, Ahmad H. Utilisation of apple pomace as livestock feed: A review. Indian Journal of Small Ruminants. 2015;**21**:165-179

[47] Daniel I, Sánchez CM, Martínez PE, Ronquillo RR. The use of coffee pulp as

a potential alternative supplement in ruminant diets. Journal of Agricultural Science and Technology A. 2015;**5**:214-217

[48] Barcelos AF, Paiva PCA, Pérez JRO, Santos VB, Cardoso RM. Composição química da casca e polpa desidratada de café (*Coffea arabica* L.) armazenadas em diferentes períodos. In: II Simpósio de Pesquisa dos Cafés do Brasil. 2001. pp. 818-825

[49] Mussatto SI, Dragone G, Roberto IC. Brewerso spent grain: Generation, characteristics and potential applications. Journal of Cereal Science. 2006;**43**:1-14

[50] Dewhurst RJ, Davies DR, Merry RJ. Microbial protein supply from the rumen. Animal Feed Science and Technology. 2000;**85**:1-21

[51] Daghir NJ. Poultry production in hot climates. 2nd ed. London, UK: CABI; 2008

[52] Rahim M, Alimon AR, Ivan M. Effects of fungal treatment on the *in vitro* digestion of palm kernel cake. Livestock Research for Rural Development. 2010;**22**

[53] Nuzul I. Characteristics of Malaysian palm kernel cake and its products. Journal of Oil Palm Research. 2013;**25**:245-252

[54] NRPH (National Range and Pasture Handbook), 190-vi; 1997

[55] Alimon AR. The nutritive value of palm kernel cake for animal feed. Journal of Oil Palm Research. 2004;**40**:12-16

[56] Beynen AC, Schonewille JT, Terpstra AHM. Influence of amount and type of dietary fat on plasma cholesterol concentrations in goats. Small Ruminant Research. 2000;**35**:141-147

[57] Kung L. The Role of Fiber in Ruminant Ration Formulation. Department of Animal and Food Sciences of University of Delaware Newark, Delaware, USA; 2014

[58] Mirzaei-Aghsaghali A, Maheri-SIS N. Importance of physically effective fiber in ruminant nutrition: A review. Annals of Biological Research. 2011;**2**:262-270

[59] Kaur H, Arora SP. Dietary effects on ruminant livestock reproduction with particular reference to protein. Nutrition Research Reviews. 1995;**8**:121-136

[60] Rukkwamsuk T. Effect of nutritional on reproductive performance of postparturient dairy cows in the Tropics: A review. Thai Journal of Veterinary Medical. 2011;**41**:103-107

[61] Meikle AM, Brun V, Carriquiry M, Soca P, Sosa C, Adrien ML, et al. Influences of nutrition and metabolism on reproduction of the female ruminant. In: Proceedings of the 10th International Ruminant Reproduction Symposium. 2018

[62] Bisinotto RS, Greco LF, Ribeiro ES, Martinez N, Lima FS, Staples CR, et al. Influences of nutrition and metabolism on fertility of dairy cows. Animal Reproduction. 2012;**9**:260-272

[63] Shin EK, Jeong JK, Choi IS, Kang HG, Hur TY, Jung YH, et al. Relationships among ketosis, serum metabolites, body condition, and reproductive outcomes in dairy cows. Theriogenology. 2015;**84**:252-260

[64] Nolan CJ, Neuendorff DA, Godfrey RW, Hams PG, Welsh TH, Mcarthur NH, et al. Influence of dietary energy intake on prepubertal development of Brahman bulls. Journal of Animal Science. 1990;**68**:1087-1096

[65] CBRA – Colégio Brasileiro de Reprodução Animal. Manual para exame andrológico e avaliação de sémen animal. 3 ed. Belo Horizonte; 2013.

[66] Martin-Rillo S. Bora semen evaluation in practice. Reproduction Domestics Animal. 1996;**31**:519-526

[67] Unanian MM, Silva AEDF, McManus C, Cardoso EP. Características biométricas testiculares para avaliação de touros Zubuínos da raça Nelore. Revista Brasileira de Zootecnia. 2000;**29**:136-144

[68] Soderquist L, Hulten F. Normal values for the scrotal circumference in rams of Gotlandic Breed. Reproduction in Domestic Animals. 2006;**41**:61-62

[69] Oldham CM, Adams NR, Gherardi PB, Lindsay DR, Mackintosh JB. The influence of level of feed intake on sperm-producing capacity of testicular tissue in the ram. Australian Journal of Agricultural Research. 1978;**29**:173-179

[70] Cameron AWN, Murphy PM, Oldham CM. Nutrition of rams and output of spermatozoa. Proceedings of the Australian Society of Animal Production. 1988;**17**:162-165

[71] Martinez JM, Dominguez B, Barrientos M, Canseco R, Ortega E, Lamothe C. Biometry and testicular growth influenced by nutrition on prepubertal pelibuey lambs. Online Journal of Animal Feed Research. 2012;**2**:314-321

[72] AOAC – Association of Official Analytical Chemistry. Official Methods of Analysis. 15th ed. Washington, DC: AOAC International; 1990

[73] Van Soest PJ, Robertson JB, Lewis BA. Methods for dietary fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dietary Science. 1991;**74**:3583-3597

[74] Hall MB. Neutral Detergent-soluble Carbohydrates. Gainesville: Nutritional Relevance and Analysis; 2000

[75] Silva DJ, Queiroz AC. Análise de alimentos: métodos químicos e biológicos. 3a ed. Viçosa, MG: UFV; 2002

[76] NRC – National Research Council. Nutrient Requirements of Dairy Cattle. Washington, DC: National Academy of Science; 2001

[77] Martin GB, Blache D, Miller DW, Vercoe PE. Interactions between nutrition and reproduction in the management of the mature male ruminant. Animal. 2010;**4**:1214-1226

[78] Santos AX, Kahwage PR, Faturi C, Quinzeiro Neto T, Lourenço Júnior JB, Joele MRSP, et al. Feed supplementation with palm kernel cake-based concentrate increases the quality of water buffalo semen. Animal Reproduction. 2014;**11**:85-95

[79] Rege JO, Toe F, Mukasa-Mugerwa E, Tembely S, Anindo D, Baker RL, et al. Reproductive characteristics of Ethiopian highland sheep. II Genetic parameters of semen characteristics and their relationship with testicular measurements in ram lambs. Small Ruminant Research. 2000;**37**:173-187

[80] Cheah Y, Yang W. Functions of essential nutrition for high quality spermatogenesis. Advanced in Bioscience and Biotechnology. 2011;**2**:182-197

[81] Azizunnesa BF, Bari FY, Alam MGS. Effects of concentrate supplementation on reproductive performances and semen quality of indigenous rams in Bangladesh. Journal of Embryo Transfer. 2013;**28**:325-335

[82] El-Azab AI, Khadrb NA, Zahran K. Effect of non-protein nitrogen in the ration on semen quality. Small Ruminant Research. 1998;**27**:73-77

Section 2

Fish Farming

Chapter 5

Nile Tilapia "Oreochromis niloticus" Farming in Fresh and Geothermal Waters in Tunisia: A Comparative Study

Sami Mili, Rym Ennouri, Manel Fatnassi, Hajer Zarrouk, Rabeb Thabet and Houcine Laouar

Abstract

This work aims to compare the farming of Nile Tilapia *Oreochromis niloticus* in fresh and geothermal waters through monitoring the specie's zootechnical parameters: growth, mortality and feed conversion rate. For geothermal water rearing, fish was placed in cages in Bechima Station, in southern Tunisia, while Smati Reservoir, in the center of the country was used for fresh water. The spawners were first adapted to geothermal waters in Bechima experimental station. Then, the broodstock phase lasted 60 days and allowed the obtainment of 1–2 g larvae. Fertility was important and varied between 451 and 1589 larvae/female, which is associated with the females' total weight ($F = 1.6\ W^{2.1}$). In the pre-growing phase, the comparison of fry growth rates (weight 1.3 g) in the geothermal and freshwaters showed a small variation with recorded rates slightly in favor of fish bred in fresh water. During 50 days within the breeding phase, fish weight achieved in freshwater was more important reaching 12.7 g (TCJ = 0.228 g /day compared to 10.51 g (TCJ = 0.184 g/day) recorded in geothermal waters. Similarly, during the fattening phase, the weights gained after 30days demonstrated better growth rates for tilapia cultured in freshwater (up to 60 g) in contrast to that bred in geothermal water (35–40 g).

Keywords: *Oreochromis niloticus*, farming, freshwater, geothermal waters, Tunisia

1. Introduction

Across the ages, Man has been harnessing water resources to suit his needs whether for power generation or for food, fishing and aquaculture. The first aquaculture tryouts started more than 4000 years ago in Egypt with the production of the famous fish: Tilapia [1]. Till to date, tilapia has remained an important food resource, in the family Cichlidae, with two species being predominantly cultivated: Mozambique Tilapia (*Oreochromis mosambicus*) and Nile Tilapia (*O. niloticus*), which alone

IntechOpen

accounts for more than 1/5 of the total global aquaculture production; and 85% of tilapia production [1–4]. Nile tilapia, a species native to the Nile River, also inhabits the Niger, Volta, and Senegal River Basins [1, 5, 6]. Thanks to its suitability for aquaculture and given its quick adjustment to extreme and varied environments, its ease of reproduction in captivity and its wide ecological valence, this fish has been the subject of various breeding attempts and has been widely spread in all continents since the 1960s [7, 8].

Around the world, several methods have been used to breed tilapia, including extensive (mostly in ponds), semi-intensive, intensive and mostly hyper-intensive systems. These methods are characterized by the use of selected strains and a high-performance compound feed [4, 9]. Nile Tilapia is widely known for its adaptability to different farming systems linked to its spawning period which spreads over the whole year. Likewise, this species is known for its resistance to pathogens as well as its ability to withstand stressful environments and the different manipulations associated with aquaculture [10].

In addition to the afore-mentioned advantages, *O. niloticus* has an intriguing growth rate, unique when compared to other species of the cichlid family. On top of a good food conversion ratio associated with an excellent ability to accept artificial feed [11–13], its diet corresponds to the lowest levels of the food chain (phytoplankton, detritus...). All these attributes make the production costs of this species relatively moderate and adequate. In Tunisia, in addition to mullet, pikeperch and carp, the rearing of *O. niloticus* could prove to be an interesting future alternative for the partial replacement of the production of other freshwater fish, given its very remarkable biological characteristics in inland fish farming.

Tilapia was first introduced in 1966 in Kebilli Oases, where aquaculture is often connected with agricultural practices, taking advantage of the warm climate and the abundant geothermal water resources in southern Tunisia. However, despite the unerring documentation of Tilipia's survival and reproduction [14], it was not until 1999, that the idea of breeding this species was pursued. This took place after the experimental research station in geothermal pisciculture of Bechima in the governorate of Gabes was founded by the National Institute of Sciences and Technologies of the Sea (NISTS). Since then, several experiments have reported effective management and success of the different rearing cycles of this species, from spawning to grow-out [15]. In parallel, various acclimatization experiments of tilapia have been conducted in the southern oases, as well as in the reservoirs in the north and center of Tunisia, where the low winter temperature hinders the growth and survival of this species. These experiments have shown encouraging results and breeding success especially at the reservoirs of Lebna, Lahma, Ghezala and Sidi Saâd [16, 17].

The present study was commissioned by the Technical Center of Aquaculture (TCA) and the Higher Institute of Fishery and Aquaculture of Bizerte (HIFAB), within the framework of a convention signed in 2015 between the TCA and the NISTS which aims to ensure the annual production of 300,000 single-sex male Nile tilapia fry. In other respects, the researchers sought to identify the most suitable environment for the rearing of this species in Tunisia through pinpointing the strengths and weaknesses associated with this species' farming in two quite different environments (fresh and geothermal waters). To achieve this goal, the physicochemical parameters of the study areas, zootechnical parameters of the fish and pathologies were considered on top of the monitoring and evaluation of the different rearing phases (hatching, larval rearing, and grow-out) and the economic study.

2. General overview of the hydric and aquaculture potentials in Tunisia

2.1 Aquaculture in Tunisia

In Tunisia, according to the General Directorate of the Environment and Quality of Life [18], 5 types of aquatic livestock farming activities currently exist: marine fish farming, inland fish farming, fish farming in geothermal waters, shellfish farming and tuna grow-out. These aquaculture activities date back to the 1960s and they started with the rearing of sea bass *Disentrarchus labrax* and sea bream *Sparus aurata* in marine waters. At that time, the competent authorities set up a strategy for the exploitation of the dams' reservoirs as a significant hydro-ichthyic support. However, the first attempts to introduce freshwater species into the reservoirs have proven that only carp (*Cyprinus carpio communis*, *Cyprinus carpio specularis*, *C. carpio coriaceus*), roach (*Rutilus rubilio*) rudd (*Scardinius erythrophtalmus*), pikeperch (*Sander lucioperca*), black-bass (*Micropterus salmoides*), mullets (*Chelon ramada*, *Mugil cephalus*) and Nile Tilapia (*O. niloticus*) were able to acclimatize [14, 17, 19]. That said, inland fish farming in water reservoirs and hill lakes in Tunisia remains of paramount importance and contributes with 1000 tons/year in the total fishery production [20].

2.2 Water potential and aquaculture sector in Tunisia

According to the GDDLHW [21], the Tunisian water network extends over a total area estimated at 20,000 ha, retaining more than 4.2 billion m^3 of water. This network consists mainly of dams, hill reservoirs and hill lakes generally located in the north and center of Tunisia. Eventually, the possibilities of development of the aquaculture sector are quite considerable. Indeed, Tunisia has significant natural potentials from north to south, distinguished by the presence of about 30 reservoirs and hill lakes, spread over 13 governorates between the north and center of the country. In addition, the south offers the opportunity to integrate fish farming with agriculture, where groundwater can efficiently contribute to the diversification and production of certain species in geothermal waters including Nile Tilapia. In fact, a great variety of hot water resources exists in southern Tunisia including the geothermal waters whose exploitable quantity is estimated at 737.8 Mm^3/year [13]. This potential consists of deep resources mainly distributed among three aquifers: Intercalary Continental, terminal complex aquifers and Jefarra Basin [15]. This prospect helped initiate endeavors for both tilapia farming and agriculture. Indeed, the idea has converged towards the diversification of breeding species with respect to those introduced in the reservoirs (mullet, pike perch, catfish). Thus, a twofold exploitation and valorization of geothermal water has been achieved as the same water is used for both agriculture and fish farming enriching the agricultural areas with nutrients and fish organic elements [15]. However, the type, accessibility and quantity of the water available, can constitute significant limiting factors for the initiation of tilapia production in the south [16].

3. Overview on Tilapia production

3.1 World production

In 2015, Tilapia group ranked third in terms of production worldwide after Cyprinidae and Salmonidae. Global tilapia production has increased rapidly since the

1980s and reached 3,670,259 tons in 2014 [22]. This production is mainly destined to the U.S. market, the main importer of this fish. In terms of geographical location, Asia accounts for more than 80% of tilapia production in the world, with China as the largest producer with 1 M tons. On the other hand, while Africa is the native environment of tilapia, its production remains extremely limited (apart from Egypt and Zimbabwe) [22, 23].

3.2 Tilapia production in Tunisia

Nile tilapia (*O. niloticus*) was the first exotic species reared in Tunisian geothermal waters, particularly due to its rapid growth and tolerance to high temperatures. In addition, the remarkable adaptation of this species and the highly motivating results of its breeding have attracted the private sector to invest in this activity since 2009 in the governorate of Medenine [20]. However, the production of tilapia has remained low compared to other groups of freshwater fish (5428 Tons against 1,034,035 compared to the rest of the species). Most of this production is realized in the reservoirs of Lakhmés (Siliana), Lebna (Nabeul) and Sidi Saâd (Kairouan) [20]. The average market price is also low (3–4 TD/kg; which corresponds to 1.2–1.7 €) and remains uncompetitive compared to other farmed fish. The reason is that tilapia, like most freshwater fish, is not in the dietary habits of Tunisians and its market is quite small.

4. Presentation of the species *O. niloticus* (Linneaus, 1758)

Like all tilapias, *O. niloticus is a* member of the family Cichlidae (**Figure 1**). The systematic position adopted for this species is the following [1, 9] Phylum: Vertebrates; Subphylum: Gnathostomes; Superclass: Fishes; Class: Osteichthyes; Subclass: Teleosts; Order: Perciformes; Suborder: Percoids; Family: Cichlidae; Subfamily: Tilapinae; Genus: *Oreochromis*; Species: *niloticus*.

4.1 Morphological characteristics of the species

Nile tilapia is a Cichlid belonging to the group of maternal mouth-brooder fish. It is characterized by a grayish coloration with pinkish breast and flanks, alternating light, black vertical stripes clearly visible especially on the caudal fin and the posterior part of the dorsal fin, a high number of long and thin gill rakers (18–28 on the lower part of the first gill arch and 4–7 on the upper part), a long dorsal fin with a spiny anterior

Figure 1.
Nile tilapia (present work).

part (17–18 spines) and a soft posterior part (12–14 rays), and a black border on the dorsal and caudal fin in males [24–26]. *Oreochromis niloticus* can be easily distinguished from any other species of tilapia that has more or less the same characteristics mentioned above, i.e., *O. aureus*, but which shows in males an additional red border along the edge of the dorsal and caudal fins [27, 28].

4.2 Ecological requirements

Thanks to its high adaptability to biotic and abiotic ecological factors, Tilapia rearing can be carried out in fresh and warm waters, as well as in well-controlled conditions after acclimation [17]. *O. niloticus* is a eurylecious species that adapts to large variations of ecological factors as it is able to adjust to extremely different environments [29, 30].

Table 1, below, summarizes the various average values of the physicochemical parameters that this fish tolerates for its survival.

4.2.1 Temperature

Temperature represents a main factor that conditions either way the properties of the water required for rearing and the different growth phases of tilapia. In natural environment, tilapia is a eurythermic fish that can withstand important temperature disparities [32]. Thus, it is possible to find this fish at temperatures ranging between 14 and 33°C. Within breeding conditions, the lower and upper lethal temperatures recorded are 7.4 and 40.73°C respectively [33, 34]. Above 16–17°C, the fish stops feeding and becomes increasingly susceptible to a series of diseases [35–38]. For reproduction, the appropriate temperature ranges between 22 and 30°C [39–41]. In Tunisia, the optimal average temperature for tilapia breeding is between 9 and 28°C in the reservoirs. Moreover, Derouiche et al. [17] reported that this species can adopt a hibernation strategy to survive and grow when the winter temperature is around 9°C (Lebna Reservoir). On the other hand, conducted experiments of the species farming in southern geothermal groundwater show a tolerance of 36 and 40°C [42, 43].

4.2.2 Salinity

Although most tilapia are freshwater species, their ability to adapt to different salinities is clearly remarkable [44, 45]. For example, *O. niloticus* can adjust to waters with salinity between 0.015 and 30 PSU [46]. Similarly, in Tunisian geothermal waters, tilapias show their ability to withstand high salinities up to 28 g/l [43]. However, with regard to reproduction, this fish would be unable to reproduce in a salinity that exceeds 15–18‰ [47, 48].

Parameter	T (°C)	Salinity (PSU)	Alkalinity (mg/L)	Hardness (mg/L)	Ammonia (mg/L)	Dissolved oxygen (mg/L)	pH
Interval	26–32	0–20	> 20	< 50	< 0.1	3–5	6.5–8.5

Table 1.
Water quality requirements for Nile Tilapia culture [31].

4.2.3 Dissolved oxygen

Tilapia are capable of surviving in conditions where the dissolved oxygen concentration is very low [49]. Indeed, they can even withstand levels below 0.5 mg/l, which is considered to be below the threshold level tolerated by most aquaculture species [50, 51]. However, a minimum level of 2 to 3 mg/l is recommended in rearing, otherwise, a depression of the metabolic rate and growth can affect the production.

4.2.4 The hydrogen potential (pH)

Nile Tilapia presents a capacity of survival in environments of extreme pH. However, the optimal pH advised for its survival and its breeding oscillates between 7 and 8 [39, 45, 49].

4.2.5 Nitrogenous compounds

In fish farming, ammonia poisoning is closely related to pH. When this substance increases, it leads to the transformation of a significant amount of total ammonia into its toxic form (NH_3) [16, 52]. The concentration of nitrogenous metabolic waste excreted through the gills, urine depends mainly on the temperature, the size of the individuals, and the quantity and amount of food distributed. This concentration must be kept below the critical threshold of *O. niloticus*, not exceeding 5 mg/l for nitrate, 500 mg/l for nitrite, and 15 mg/l for total ammonia [53].

4.2.6 Photoperiod

The action of light, although closely related to temperature, acts on the species' growth via the endocrine system. Mélard [54] explain that an optimal photoperiod stimulates the secretion of growth hormone (GH) in *O. niloticus.* Moreover, larvae are more sensitive to photoperiod than fry and juveniles [48, 55]. Experimentally, larvae that are exposed to a long period of light (18-24 h) have a better growth and a significantly higher food efficiency than those exposed to a short or intermediate period between 6 and 12 h [55].

4.3 Reproductive biology

4.3.1 Reproductive behavior

In the wild, when abiotic conditions are appropriate, adults migrate to a shallow area with a loose substrate (gravel, sand, clay). After choosing the site for their own nest, each male aggressively defends its territory and digs a plate-shaped nest with its mouth. The females living in a school near the breeding grounds move between the males and each one tries to acquire a partner [15, 56].

4.3.2 Sexual maturity

The size of the first sexual maturity of *O. niloticus* varies between 14 and 20 cm. However, under stressful conditions, this species can reproduce as early as 3 months of age, at a weight of less than 50 g [57]. Moreover, the reproduction period of this species is exponentially continuous during the whole year when the water temperature is higher than 22°C [52]. Thus, in Tunisia, the study conducted by Azaza et al. [58] on the reproduction of Nile tilapia in captivity in the geothermal waters in southern Tunisia, showed that this fish reaches its first sexual maturity during the first year of rearing, with an Lm50 equal to 11.3 cm for females and 12.3 cm for males.

4.3.3 Fertility

Absolute fecundity is defined as the number of eggs freshly recovered from the oral cavity of a female. In tilapia, as in other fish, this fecundity increases with the size of the females. As reported by Mélard [54], the minimum absolute observed fecundity for a 26 g female is 340 ovules and the maximum fecundity for a 550 g female is 3500 ovules. In addition, Dhraief et al. [8] proved that this parameter increases with the length of the females. On the other hand, Mélard [54] demonstrated that relative fecundity (expressed as the number of fertilized eggs or fry produced/kg of female) varies inversely with the average weight of tilapia females.

4.4 Growth

It is commonly accepted that Fish have a predetermined typical growing course dependent on genetic factors which interact with other environmental aspects. Thus, the growth rate is extremely alterable depending on the controlling factors, such as temperature and limiting factors including food, oxygen, and ammonia which affect the amount of energy available for growth. Similarly, other secondary factors such as stocking density and photoperiod, can certainly affect the growth of the species [59]. Moreover, in *O. niloticus* there is a phenomenon of sexual dimorphism of growth which appears very quickly in rearing: the males have better growth performances than females, due to the particularity of the reproductive process in females (oral incubation) and social behavior (territoriality, etc.) [24].

4.5 Production of single-sex male population

In order to optimize *O. niloticus* production systems, rearing of single-sex male populations is sought more and more in tilapia farming for many reasons. First, males grow twice as fast as females [35, 60]. Another reason is to avoid reproduction which would result in an overpopulation of small individuals in the rearing environment [61], and eventually ensure a homogeneous population at the time of harvest, with an interesting individual size and good commercial value.

4.6 Pathological risks

Like all aquatic species, Nile tilapia can be prone to a range of diseases resulting from the proliferation of certain pathogenic organisms. Generally, bacteriological diseases remain the most prevalent, namely Mobile *Aeromonas septicemia* and *Vibriosis*, resulting primarily from stress and poor water quality. Affected fish show signs of burns on the skin and fins and a loss of balance associated with abnormal behavior [9, 52]. Results obtained from a study done in tilapia farms in Ghana, revealed three types of ectoparasites: *Trichodina sp*, *Monogenes* and *Tetrahymena sp*, of which the first two were common in most farms, but did not pose real problems.

4.7 Diet

In the wild, tilapia is an omnivorous fish. In aquaculture, however, it shows an ability to consume various products, co-products and waste products that are considered valuable, such as palm nuts, soybean or cotton cakes, rice flour, rapeseed, alfalfa and animal excrements [51, 62]. In Tunisia, a study conducted on the development of dry feeds for *O. niloticus* by Derouiche *et al.* [17] showed that the best growth and feed conversion rates were obtained by feeds containing 20% and 30% of fish meal, with conversion rates of 1.71 and 1.49.

5. Materials and methods

5.1 Presentation of the study areas and breeding structures

The aim of this work is to identify the most suitable environment for the rearing of *O. niloticus* in Tunisia. To achieve this goal, this study was held investigating two areas with different types of waters: the experimental station of Bechima (geothermal water) and Smati Reservoir (fresh water). In addition to the main infrastructure intended for the preparation of broodstock for reproduction, spawning and the production of larvae, the production of juveniles and young individuals requires appropriate infrastructure, such as cages and enclosures which are efficient and resistant to environmental factors.

5.1.1 Bechima experimental station

Bechima experimental station is a research unit created in 1999 by the NISTS in Al-Hamma region in the governorate of Gabes in southern Tunisia. It is located in the vicinity of a cooler on a slope of 3% to ensure a continuous circulation of water by simple gravity. Two artesian wells supply the station with geothermal water at a temperature of 70°C. This water is cooled by the atmosphere to reach an average temperature of about 30–40°C. In terms of infrastructure, the station is equipped with 3 greenhouses containing breeding, larval rearing, hatching and rearing tanks [15].

5.1.2 Smati reservoir

Smati Reservoir covers an area of 121 km^2 and is located in the region of Al-Ala in the governorate of Kairouan, central Tunisia on Smati Wadi. The average salinity and depth are estimated at 2.3 PSU and 1.5–2 m respectively. In addition, according to the General Directorate of Fisheries and Aquaculture [20], this reservoir offers ichthyic habitat for two species namely the mullet *M. cephalus* and the Barbel *Barbus callensis*, with a total production estimated at 17.3 tons in 2015.

5.1.3 Design and assembly of breeding structures

In order to facilitate handling and transport and to minimize costs, the used cages and enclosures were built with respective volumes of 3 m^3 (with Length = 1.5 m, Width = 1.3 m and Height = 1.5 m), and 2 m^3 (with Length = 1.3 m, Width = 1 m and Height = 1.5 m), using a mesh size of 6 mm of polyamide nets. In total, 31 cages were constructed, of which only five were initially installed in the large tank for the phases of pre-growing and grow-out in Bechima Station. In addition, six enclosures were built for the rearing of individuals in a semi-extensive system in Smati Reservoir, of which two were used for hatching and two others for rearing at a depth of 1.5-2 m [63].

5.2 Biological material and breeding procedure

5.2.1 Biological materials

O. niloticus, in particular the strain "Maryout", subject of this zootechnical study was introduced in 1999 from Libya within the framework of research cooperation between the NISTS and the Marine Science Center of Tajoura [64].

The experiments started with the collection of more than 6000 larvae (average weight = 0.01 g) from 13 broodstock (weight between 99 and 190 g) which were well adapted to life in captivity in the geothermal waters of Bechima. These larvae were subdivided into 2 batches, to be prepared for an initial pre-growing phase of 60 days in two larval tanks, located in the larval rearing greenhouse of the station, with the aim to obtain fry of 1 to 2 g. The latter were then transferred to two cages and two enclosures which were installed respectively in Bechima Basin and in Smati Reservoir in order to obtain juveniles of 15–20 g (50 days). Finally, a sorting was ensured based on average weight and sex to produce neo males and to start the last phase of production: the grow-out.

5.2.2 Breeding procedure

5.2.2.1 Collection of eggs and larvae

During this study, the broodstocks were maintained under optimal abiotic rearing conditions. In fact, several signs allow the identification of incubating females, such as the appearance of a dark band on the forehead and black spots on the flanks, as well as a fast and discontinuous swimming with a rather aggressive behavior towards other specimens in the tank.

5.2.2.2 Females reproductive parameters

Once the female identified and isolated, they are weighed and measured. This procedure allows the evaluation of two important parameters: relative and absolute fecundity, which describe the productivity of the females per day according to weight and length. The absolute fecundity refers to the total number of eggs present in a female before fertilization. Relative productivity represents the total number of larvae produced per day in relation to the total biomass of females in g.

5.2.2.3 The broodstock phase

Larvae were obtained by spitting technique. The handling of the larvae as a count is a delicate operation, requiring a particular follow-up not to stress the unhatched eggs and larvae. The latter were reared for 60 days in cubic tanks with a unit volume of 1m^3, a density of 800 larvae/m^3, and a flow rate of 4 to 6 l/min. As for nutrition, larvae were fed 40% of protein levels in order to obtain fry with an average weight of 1 to 2 g.

5.2.2.4 The breeding phase

The breeding/pre-growing phase started on May 11th, 2017, in the floating cages installed in the rearing tank at Bechima experimental station (average weight: 1 to 2.03 g). One day later, 3009 larvae (average weight of 1.35 g) were transferred to the enclosures. At the end of this phase, a sexual sorting of the fry was required to start the rearing of the mono-sex populations. The density adopted for each structure is 500 individuals/m^3. Moreover, beyond a size of 20 g, sexual dimorphism can be observed and the distinction between males and females is possible. Indeed, in males, the genital papilla is protuberant in the shape of a cone and carries a urogenital pore at the end. On the other hand, in females, it is small and round, found in the middle of a transverse slit (oviduct) which is located between the anus and the urethral orifice (urinary pore) located at the end.

5.2.2.5 The fattening phase

From mid-July onwards, we started the fattening/grow-out phase of the reared fry. In general, the sorting was first based on size that allowed the retainment of individuals with an average weight greater than or equal to 20 g. Towards the end of August, males and females were reared separately to later evaluate growth rates for both sexes. Regarding nutrition, the supplemented food for tilapia was the same in both environments with 30% of protein level and was distributed at a rate of 10% on a daily basis.

5.2.2.6 Watching out for pathology

In order to ensure the success of the different rearing stages, a certain level of cleanliness and hygiene in the experimental tanks had to be maintained on a daily basis. The bottom was siphoned off before feeding, in order to eliminate feces and any kind of deposit of food, thus avoiding the development of pathogenic organisms. In addition, specific measures related to the health of the group, were taken while monitoring the condition of individuals and their behavior.

5.2.3 Nutrition

The added food was composed of raw materials available on local market and prepared in Bechima pilot station, with variable proportions according to the fish stage development and following the formulas provided by the research of the NISTS (**Table 2**).

Ingredient (%)	Broodstock	Larvae	Fry	Adults
Soybean cake	41	45	50	43
Fishmeal	14	25	20	14
Corn	35	20	20	935
Vegetable oil	8	8	8	6
VMS	2	2	2	2

Table 2.
Composition of tilapia food according to rearing phases [13].

This food is rich in protein with a level up to 40%. In addition, vitamins and minerals from the Mineral Vitamin Supplement (MVC) were provided in the food to meet the dietary requirements of the species. The rationing rate, the granulometry and the quantity of the food supplied vary according to the development stages, size and age of the individuals, the energetic value of the food and the variation of the physicochemical properties. The adopted rationing rate ranges from 20 to 10% (from the first day until pre-growth). The amount of food for broodstock (Qg) and for larval rearing (Ql) was determined in accordance with the following formulas:

$$\mathrm{Qg} = (\mathrm{Nm} * \mathrm{Mm} + \mathrm{Nf} * \mathrm{Mf}) * \mathrm{TC} \tag{1}$$

$$\mathrm{Ql} = (\mathrm{Bf} - \mathrm{Bi}) * \mathrm{TC} \tag{2}$$

With: Nm and Nf: numbers of male and female spawners; Mm and Mf: average weights of male and female spawners; TC: conversion rate; Fb and Bi: final and initial biomass during the larval stage.

Additionally, the remaining, not ingested, amount of food was monitored and estimated in order to fully adjust the supplemented amount to the needs of the fish.

5.2.4 Zootechnical parameters

To estimate the growth of fish during the different rearing phases, a number of indices and zootechnical parameters should be calculated.

5.2.4.1 Body weight gain

This index is used to evaluate the weight growth of fish during a given time. It is calculated according to the following formula:

$$\text{Gain } \textit{in} \text{ average weight (g)} = \text{final weight (g)} - \text{initial weight (g)} \tag{3}$$

5.2.4.2 Daily growth rate (DGR)

This parameter is determined for a short period of time from a fish sample during all rearing phases and is estimated according to the formula:

$$DGR = \frac{Final\ Mass - Initial\ Mass}{Time} * 100 \tag{4}$$

5.2.4.3 Mortality

This zootechnical parameter is of crucial importance when evaluating the rearing performance. It is therefore vital to monitor the cumulative mortality rate in the larval, fry and juvenile fish stock. This is the ratio of the number of dead individuals to the total population during a given timespan.

$$Mortality\ rate = \left(\frac{Nomber\ of\ dead\ organisms}{Total\ Nombr}\right) * 100 \tag{5}$$

5.2.4.4 Survival rate

The survival rate is well correlated to the mortality rate. It is calculated from the total number of fish at the end of the experiment and the number of fish at the beginning of the rearing, according to the following relationship:

$$Survival\ Rate = \left(\frac{Final\ specimens\ number}{Initial\ specimens\ number}\right) * 100 \tag{6}$$

5.2.4.5 Food conversion rate (FCR)

It is a food conversion index that measures the efficiency of the transformation of nutrition into fish flesh. It represents the ratio between the total amount of the supplied food to the fish and the gain obtained in fish weight.

$$\mathrm{FCR} = \frac{\text{Dry weight of the supplied food}}{\text{Gain in fish weight}} \tag{7}$$

5.2.4.6 Daily feeding ration (DFR)

It is the ration supplied per day of rearing, and it depends closely on the feeding rate.

$$DFR = ((Average\ weight * Feeding\ rate)/100)) * Nombr\ of\ larvae \tag{8}$$

5.2.4.7 Length-weight relationship

In order to properly control the growth parameters of broodstock, pre-grown fry and grown individuals in both environments, it is necessary to establish the relationship between the size and weight of the fish, which is defined according to Le Cren [65] by the following equation:

$$\mathrm{W} = \mathrm{aL^b} \tag{9}$$

with W: the weight of the fish in grams; L: the length of the fish (Tl, Fl or Sl) in centimeters; a: slope; b: coefficient of allometry defined as the coefficient of relative growth in weight.

Three cases can be distinguished: if b = b theoretical, there is an isometric allometry between the two characters, if b $<$ b theoretical there is a negative allometry, and if b $>$ b theoretical, the allometry is positive [66].

5.2.5 Monitoring of physico-chemical parameters

Throughout the whole experiment and during all the rearing phases, monitoring the physicochemical parameters was of primary concern. The different parameters, notably temperature, salinity, pH, and dissolved oxygen were measured with a Multi 350i/SET. Nitrites, nitrates and ammonium were also kept track of, because of their impact on water quality. The various physicochemical analyses were made in situ and in the laboratory.

5.2.6 Data analyses

The study of growth is a very delicate approach in fisheries and it requires a method which best suits the basic data, and the choice of the model that effectively describes the relationship between the variables. For this reason, and in order to have solid understanding of the prior effects of physicochemical and zootechnical parameters on the success of the rearing, it is necessary to carry out some statistical tests based on R, in order to determine the most suitable environment for tilapia farming. Thus, tests were conducted to determine the effect of different physicochemical parameters on larval rearing, pre-growth and grow-out.

6. Results and discussion

6.1 Evaluation of the rearing parameters of *O. niloticus*

6.1.1 Evaluation of broodstock fecundity

In this study, broodstocks were maintained under optimal abiotic conditions, thus 13 females ranging in size from 99 to 183 g were collected in the post-spawning phase. We established relationships linking the number of larvae obtained to the size and females body weight in order to study the absolute fecundity. In addition, the data were fitted with power curves, whose equation is $y = aX^b$.

The absolute fecundity observed during the first cycle varied between 451 and 1598 larvae/female (L/F), for respective weights of 113.4 and 183 g. However, the calculated average fecundity (806 L/F) was highly significant compared to research made by NISTS (600 L/F) and TCA (510 L/F) [30, 67]. The study of different parameters that can have a direct effect on fertility allowed us to establish the following relationships: height/weight, height/fertility and weight/fertility.

The analysis between the studied parameters shows a strong correlation between the size or total weight and the number of larvae produced by a female (R^2 = 0.6 and 0.645) (**Figure 2**).

The relationship between total tilapia broodstock weight and total length can be expressed as follows: $Pt = 0.0324\ Lt^{2.7504}$ (R^2 = 0.88) (**Figure 3**).

With regard to the allometry coefficient, for all the broodstock exploited during the first rearing phase, it was below 3 (b = 2.75 < 3) indicating that allometry is negative for this species. This result affirm that *O. noloticus* gains in length more than it does in weight. Our results are concordant with those found by Coulibaly [68] at Lake Volta in Burkina Faso, but different from those obtained by Derouiche *et al.* [17] at Lebna Tunisian Reservoir (isometric allometry), and Du Feu [69] at Lake Kainji in Nigeria (positive allometry).

Mélard [54] observed variability in fecundity for the same size. The fecundity increases significantly as a consequence of the length of the females. These findings are similar to other results achieved by authors who studied Nile tilapia in Tunisia and demonstrated that b oscillates around 1.96 [8]. Despite the small size range in our sample (between 18.8 and 23.5), it was shown that absolute fecundity evolves proportionally with size, which is congruent with results obtained by other authors [15, 54, 70]. Generally, this is largely attributed to a genetic difference, and a possible complex interaction between fecundity, egg size obtained and the staggered periodicity of egg laying.

In contrast, for the same size range, fecundity was much lower in the *Oreochromis niloticus* buccal incubator compared to substrate-laying tilapia. These species fecundity

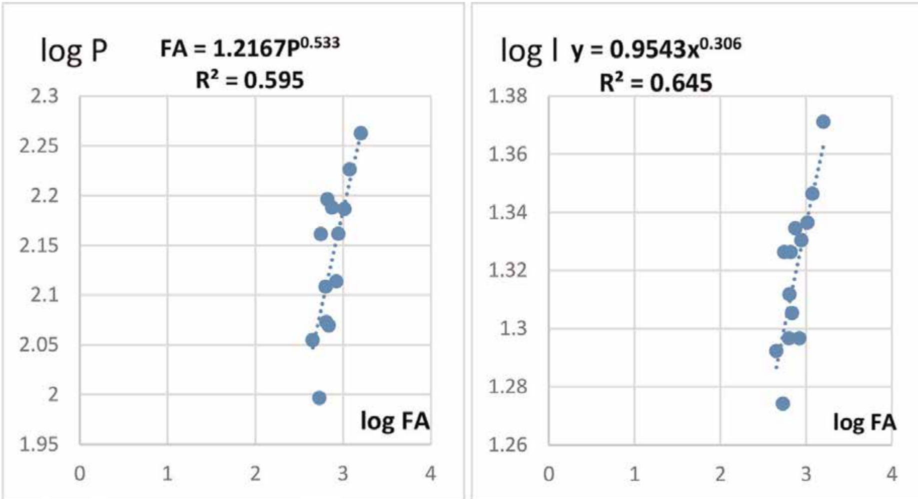

Figure 2.
Relative growth and fecundity curves for Oreochromis niloticus *broodstock.*

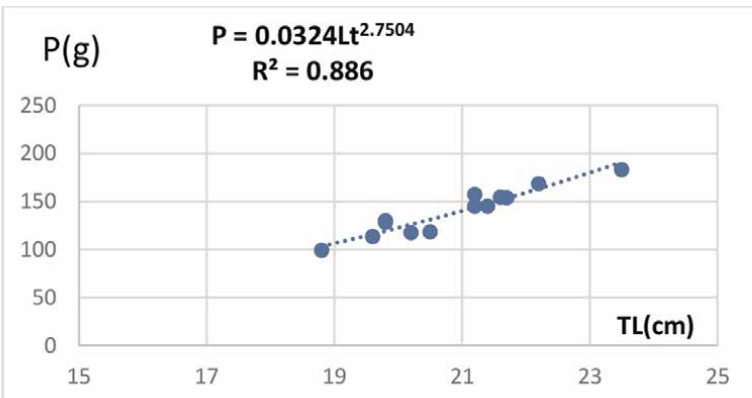

Figure 3.
Growth curve of female Tilapia.

ranged from 2314 to 5178 eggs in *Tilapia zillii* [71]. On the other hand, the results obtained on absolute and relative productivity are in close agreement with the research results achieved by Dhraief et al. [8]. In addition, the estimated relative productivity, which evolves significantly as a function of total female weight (2.43 to 10.08 larvae/g female), remains significant compared to that found in 2015 by TCA [30] (between 0.5 and 4.02) and Dhraief et al. [8] and which ranges between 1.4 and 6.8 larvae/g female.

6.1.2 Growth during broodstock phase

6.1.2.1 Evaluation of zootechnical parameters

Monitoring the evolution of the average weight of larvae, from the first phase, for 60 days allowed an estimation of pre-growth rates. A total of 6138 *O. niloticus* larvae, with an average weight of 0.01 g, were collected from 13 broodstocks on April 5th, 2017.

After 36 days, all the 0.2–0.4 g larvae were distributed to two tanks with a volume of $1m^3$. The average weight of the larvae in the first batch increased from 0.4 g (initial weight: day of collection) to 2.55 g at the end of the experiment, which corresponds to a growth rate of 0.059 g/day. For the second batch, the larvae monitoring showed a growth rate about 0.023 g/day. This rate is considerably lower than that of batch 1 whose average weight evolved from 0.01 g to 0.85 g. The evolution of the larvae weight from the first phase of rearing which lasted 60 days allowed to estimate the growth rates. These are shown in **Figure 4**.

During this period, larvae were fed manually at a rate of 2 to 3 meals per day with ground powdered food consisting of fish meal, soybean, corn, vegetable oil and a mineral-vitamin complex. The product is fed slightly in excess with a feeding rate of 20% of the total larval biomass. The initial daily intake is 7 g of food/day. Regarding the mortality, we noted that at the end of the larval rearing cycle, the survival rate was about 70%. This rate is comparable with that obtained by the NISTS (in geothermal waters) and the TCA (in fresh waters). **Table 3** summarizes the different parameters, estimated during the larval rearing phase, obtained in previous studies.

Towards the end of larval rearing, the average recorded weight of larvae ranged from 0.85 to 2.55 g between the tail and head in the batches, respectively. Our results are slightly above the individual weights estimated by NISTS and TCA [30, 67].

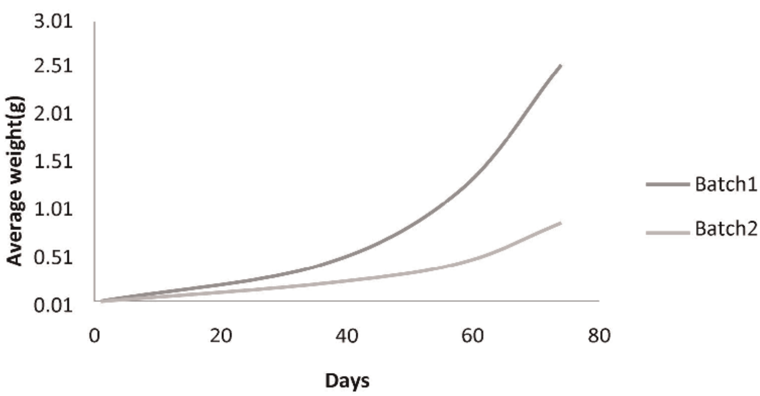

Figure 4.
Evolution of the average weight of Nile Tilapia larvae.

Breeding parameters	Bechima (Present work)	Boumhel station (CTA) [30]	INSTM [67]
Survival rate (%)	70	49.64	68–84
Duration of breeding (days)	60	40	30
Average initial weight (g)	0.01	0,012	0.012
Average final weight(g)	0.85–2.55	0.73–2.12	2.5
FCR	2.25–2.51	1.32	1.97
Average temperature	28	27	30

Table 3.
Comparative zootechnical parameters during larval rearing in Oreochromis niloticus.

The conversion index obtained in this study is 2.25 and 2.51 for batches 1 and 2 respectively. These values are higher than those attained by Philippart et al. [72] (IC = 1.1), TCA in 2015 (CI = 1.32) [30] and NISTS in 2009 (IC = 1.97) [67].

The comparison of our results with those achieved by other authors shows poor growth performance at the larval stage especially with the weight larvae heterogeneity (0.02-5 g) (size: 0.8-7 cm). This poor growth is firstly due to the: Quality and physiological state of the spawners; Egg and larval quality; Alterations during the manual counting of the larvae and during the cleaning operations of the larval rearing tanks (siphoning); Disturbance of water quality especially during the night and at dawn when oxygen is lacking; Quality of the dry food produced in the station; Low frequency of food distribution during the day (2–3 times/day), which seems to be lower than recommended (up to 8 times/day); Cannibalism phenomenon (this prompted us to make regular sorting, based on size, by separating the larvae into two batches); and High density of larvae placed in the larval rearing tanks.

6.1.2.2 Evaluation of water physicochemical parameters

The physicochemical parameters recorded in the two larval rearing tanks are relatively similar. The figure below (**Figure 5**) shows the variation of these parameters during 24 hours.

During larval rearing, the recorded temperature varied between 27.5 and 30.38 (average 28.84) during May and June 2017, respectively. To highlight the prior effect of abiotic factors on larval growth during larval rearing, statistical tests performed showed that temperature, followed by dissolved oxygen, control largely larval growth. We reported a decrease in oxygen level during the first hours of the day, which led us to install 1.5 hp. aerators to improve the performance of this parameter. On the other hand, salinity and pH seem to have a non-significant effect on larval growth.

6.1.3 Monitoring tilapia growth during rearing phase

6.1.3.1 Zootechnical parameters

As previously indicated, the fry obtained after 60 days of larval rearing was transferred to the two study areas, where the rearing and grow-out phases were initiated. The zootechnical parameters obtained in the pre-grown fry in the cages

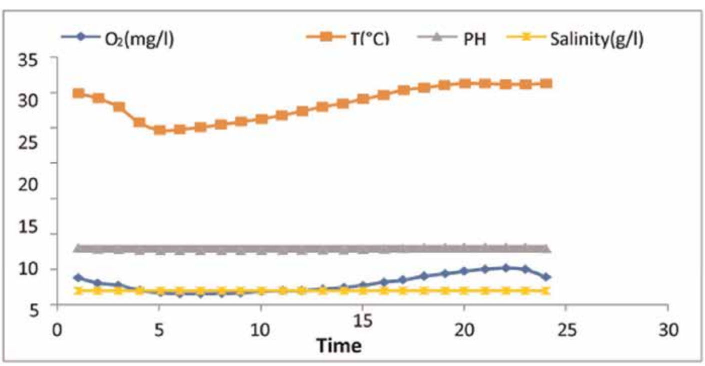

Figure 5.
Average evolution of the water physicochemical parameters.

Zootechnical parameters of breeding	Geothermal water	Fresh water
Rearing time (days)	50	50
Initial staffing	3000	3000
Final headcount	1803	1761
Mortality rate (%)	39.9	41.3
Survival rate (%)	60.1	58.7
Initial weight average (g)	1.52	1.3
Final weight average (g)	10.51	12.7
Initial biomass (g)	4545	3900
Final biomass (g)	18949.53	22364.7
Amount of supplied food (g)	25,196	24,403
Daily growth rate (g/day)	0.179	0.224
Specific growth rate (%/day)	5.98	7.13
Feed conversion ratio (FCR)	1.36	1.05

Table 4.
Zootechnical performances recorded in pre-grown tilapia in fresh and geothermal waters.

placed in the large rearing pond in Bechima Station (geothermal water) and the enclosures set in Smati Reservoir (freshwater) are reported in **Table 4**.

Growth performance expressed by the daily growth rate, the survival rate, and the conversion rate are in favor of the fish reared in the freshwater reservoir. Indeed, the growth rates recorded after 50 days of rearing are around 10.51 and 12.7 respectively for fish in geothermal water and in fresh water. Similarly, the conversion rates are of the order of 1.36 for fry reared in geothermal water and 1.05 for those reared in fresh water. For the fry length-weight relationship, we recorded a variation in *O. niloticus* growth. **Table 5** summarizes the different results obtained as well as the type of allometry.

In the freshwater pens we found that the relationship between total weight of *O. niloticus* fry and total length can be expressed as follows $Pt = 0.0096Lt^{3.1041}$

Month	Zone	Slope comparison tests			Position comparison tests			Allometry
		Tpe	(+/-)	Interpretation	Tpo	(+/-)	Interpretation	
May	ED	0.87	–	equivalents	0.235	–	Confounded	–
	EG	0.95	–	equivalents	0.89	–	Confounded	–
June	ED	1.42	–	equivalents	1.014	–	Confounded	–
	EG	1.07	–	equivalents	0.973	–	Confounded	–
July	ED	1.66	–	equivalents	1.20	–	Confounded	+
	EG	1.43	–	equivalents	1.031	–	Confounded	+

Table 5.
Tests of slopes and allometry positions of the weight-total length relationship per month in fresh and geothermal waters. (FW: fresh water; GW: geothermal water).

(n = 41, R^2 = 0.6962). On the other hand, it is expressed as $Pt = 0.0075Lt^{3.256}$ for cages placed in geothermal water. Moreover, the allometry coefficient is greater than three for the fry raised in both areas. Thus, this species exhibits a positive allometry between total weight and size. This asserts that this fish gains in weight more than in length. This result is congruent with the findings achieved at Lake Kainji in Nigeria [69] but it differs from the results found by Coulibaly [68] at Lake Volta in Burkina Faso (Minority allometry) and Derouiche et al. [17] in Lebna Reservoir (isometric allometry).

The monthly analysis of growth performance shows a slight difference in growth between the two zones over the study period. The value of the t-test is inferior to 1.96 at the 5% threshold. The month of July is characterized by a positive allometry in the reservoir and in Bechima station where the individual weight grows faster than the length.

Comparing the results of zootechnical performance between geothermal and fresh waters, it can be noted that the growth of pre-grown fry in pens at Smati Reservoir is superior to the fry raised in cages at Bechima fish farm. This may be due to the water quality at the rearing pond where the turnover rate is low. This is caused by the low flow of used water to maintain the temperature around 28–30°C. On the other hand, the water in Smati Reservoir seems to have physico-chemical parameters that are favorable to rearing.

Comparing our results with other studies, we note that at the Blonbey fish farm, fry of 1660 mg stocked in ponds at a density of 500 ind/m^3 reach an average weight of 5980 mg. The daily growth rate is 210 mg/day, while the specific growth rate is about 5.98% per day. Our results are consistent with those found by Azaza *et al.* [58]. These authors indicate that they were able to achieve a daily weight gain of 0.4666 g/day during 15 days of hatching from fry of 2 g to an average weight of 9 g. The rearing adopted by these authors was conducted on a feed with the same composition as used in our experiment.

On the other hand, FAO states that a good daily growth rate can be obtained during 30 days and under intensive conditions (0.5 g/d daily gain) [73]. In Côte d'Ivoire, conducted hatching experiments have shown that growth rates are much better in 1m^3 cages with a density of 1500 fry/m^3. These experiments produced a fry weight of 25 g during 1 month with an estimated daily growth rate of 0.22 g.

The results from the pen rearing in Smati Reservoir are similar to those achieved by Azaza et al. [58] who obtained a daily growth rate of 0.43 g/day during 45 days. However, our results differ from those found by Lazard and Legendre [59] who got an average individual weight of 5 g after 2 months of pond rearing. This experiment was based on a dry feed composed of 20% of fishmeal, for fry with an average weight of 0.9 g, which corresponds to a daily weight gain equal to 68.33 mg/day. On the other hand, the good zootechnical performance observed during our experiment in fry pre-grown in fresh and geothermal waters is comparable to that of *H. longifilis* pre-grown made during 4 weeks in 4 m^2 tanks and whose average weight evolved from 4.5 g to 50 g [74].

6.1.3.2 Evaluation of physicochemical parameters during pre-growth phase

Tilapia farming is relatively difficult because it depends on the environmental factors of the water used. Indeed, the good management of a better water quality is the key to produce fry and fish in good conditions. The average values of the physicochemical parameters recorded at the two sites during the pre-growth phase are shown in **Table 6**.

Period	05/2017		06/2017		07/2017	
	Cages EG	**Enclosure ED**	**Cages EG**	**Enclosure ED**	**Cages EG**	**Enclosure ED**
Temperature	27	25.85	27.5	24.55	35.2	28.5
Salinity (g/l)	2	0.15	2.1	0.25	2	0.2
Dissolved Oxygen	6.78	8.875	8.38	5.755	9.7	6.21
PH	8.29	7.635	8.4	7.24	8	7.32
Average weight(g)	1.3	1.51	5.17	8.2	10.5	12.7

Table 6.
Evolution of physicochemical parameters in fresh and geothermal waters.

6.1.3.3 Temperature

During the pre-growth phase, the monitoring of physico-chemical parameters in the freshwater and the geothermal water areas show that the maximum temperature values were recorded in July. The average temperature recorded in Smati Reservoir is estimated at 26.3°C against 30°C at the level of Bechima station. Eer *et al*. [75] have shown that a temperature between 20 and 30 °C is optimal for fish farming.

6.1.3.4 PH

Monitoring this parameter is momentous in farm management since inadequate pH values can influence the physiological fish state and their growth. Furthermore, it can lead to fish mortality, especially during the early developmental stages [53]. Throughout our study, pH values in the larval tanks, cages and pens showed small variations through time, but were still within the optimal tolerance range of the species (6.5–8.5) [53]. The average pH values recorded in the cages and pens are close and show a basic character of the rearing water (7.3–8.23). This species is tolerance to pH variations and it is found in waters with pH values ranging from 5 to 11; the ideal being located between 6.5 and 8.5 [53].

6.1.3.5 Dissolved oxygen

The dissolved oxygen levels measured in the two areas are within the scale of values recommended for fish farming [50]. The average oxygen level recorded varies between 6.94 and 8.28 mg/l in fresh water and geothermal water.

6.1.3.6 Salinity

Salinity monitoring showed that this parameter was almost constant during the 3 months of the experiment in both study sites. The values recorded are 2.03 g/l and 0.2 g/l respectively in the geothermal water and freshwater.

The analysis of the evolution of the physico-chemical parameters in the two areas showed that the values are well within the ranges recommended for Nile Tilapia farming. Indeed, this species is found in natural environment between 13.5–33°C [47, 76] and does not feed below 15°C [53].

In addition, *O. niloticus* tolerates both high oxygen deficits and saturations. Thus, up to 3 mg/l of dissolved oxygen, this species does not present any particular metabolic difficulty. However, below this value, respiratory stress occurs, although mortality only occurs after 6 hours of exposure to 3 mg/l. Nevertheless, this species can withstand low concentrations of dissolved oxygen for short periods. The optimum required is 5 mg/l [53].

6.1.3.7 Effect of water on tilapia growth

To test the prior effect of abiotic factors on the zootechnical performance of fish during the pre-growth phase, we established statistical tests based on general linear models (GLM), by means of quantitative variables. These tests allow the complex parametric relationships between response variable and explanatory variables to be modeled; as well as to look for the most parsimonious relationship including only the relevant variables (using the AIC selection criterion). In addition, they make it possible to test the effects of explanatory variables and their interactions.

We followed the effect of physicochemical water parameters on the evolution of the total biomass of tilapia. We found that only temperature and dissolved oxygen govern the growth of individuals reared in freshwater and geothermal waters alike. Besides, the predictive study allowed to retain the variables mentioned above, showing the lowest value of AIC.

6.1.4 Growth parameters of tilapia during the grow-out phase

6.1.4.1 Physicochemical parameters

The table below (**Table 7**) illustrates the average values recorded of the water physicochemical parameters during the fattening phase. The study made during the grow-out stage allowed us to point out a clear difference in temperature between the two study areas. For the other parameters (dissolved oxygen and pH) remained very close and did not show significant variations.

6.1.4.2 Nitrogenous elements during breeding phase

Nitrogen is an essential compound in living structures, depending on the degree of oxidation; it exists in three forms in water: nitrites (NO2-), ammonium (NH4+) and

Parameters of Study area	Temperature (°C)	Dissolved oxygen (mg/l)	Salinity (PSU)	PH
EG Bechima	33.2	7.9	2	8.25
ED Smati Reservoir	27.6	7.64	0.2	7.8

Table 7.
Variation of physicochemical parameters between Bechima Station and Smati reservoir.

Parameter Environment	Hatching				Grow-out			
	$NO2^-$ (mg/l)	$NH4^+$ (mg/l)	$NO3^-$ (mg/l)	P (mg/l)	$NO2^-$ (mg/l)	$NH4^+$ (mg/l)	$NO3^-$ (mg/l)	P (mg/l)
EG Bechima	0.05	0.27	1.4	0.11	0.07	2.13	1.19	0.32
ED Smati Reservoir	0.06	0.12	1.7	0.25	0.38	0.34	3.4	2.43

Table 8.
Evolution of nitrogenous elements for pre-growth and grow-out in the two environments.

nitrates (NO3-). The latter must be well controlled throughout the rearing period due to their toxic nature. The results of these analyses on the quality of the rearing water are reported in the following table (**Table 8**).

During the pre-growth phase, the analyses obtained in the two study areas are within the recommended ranges for rearing *O. niloticus*. In fact, the concentration of nitrogenous waste excreted by the gills and urine depends on various factors, namely temperature, fish size, ammonia concentration in the environment and the quality of the feed which must be kept below the critical threshold for survival of *O. niloticus*. Concentrations should not exceed 15 mg/l nitrate, 2 mg/l nitrite, 0.95 mg/l ammonium ions and 0.3 mg/l orthophosphate in any case [53].

However, at the grow-out phase, we recorded an increase in nitrite, nitrate and orthophosphate levels (at the reservoir). A high concentration of ammonia may have altered the taste and odor of the water. This may be explained by the increase in temperature during the summer season, and the phytoplankton bloom at the reservoir impoundment caused by the discharge of agricultural wastes and nitrogenous products. However, the majority of the waters of Tunisian reservoirs are classified as eutrophic to hypertrophic. These values remain below those recorded in the reservoir of Bir M'chergua whose nitrate concentration is estimated at 16.9 mg/l [77].

6.1.4.3 Growth parameters of Tilapia during fattening phase

The grow-out phase started in mid-July, so the monitoring period was quite restricted (30 days), and the young individuals were fed 3 times a day. Therefore, we will base our results on the daily growth rate. The following table (**Table 9**) summarizes the different zootechnical parameters recorded after 1 month of rearing.

The comparison of the growth performances between fresh and geothermal waters has shown high potentialities in favor of fish placed in the reservoir. Indeed, after 30 days, the total masses obtained varied between 30 and 59 g respectively in geothermal water and fresh water. Moreover, the highest growth rate is recorded in the

Zootechnical parameters of breeding	Grow-out in geothermal water	Grow-out in fresh water
Rearing time (days)	30	30
Initial staffing	1801	1761
Final headcount	1780	1749
Mortality rate (%)	1.17	0.69
Survival rate (%)	98.83	99.31
Average of initial weight (g)	10.4	12.7
Average of final weight (g)	30	29–59
Initial biomass (g)	18730.4	22634.7
Final biomass (g)	53,400	69,960
Amount of feed distributed (g)	16389.1	24,654
Daily growth rate (g/day)	0.65	0.54–1.54
FCR	0.47	0.2

Table 9.
Zootechnical parameters of tilapia grown in Bechima Station and Smati reservoir.

Study area	Storage density (ind/m^3)	Rearing time (days)	Initialize (g)	Finalize (g)	Daily growth rate
Sidi Saad Reservoir (Tunisia) [42]	75	75	20.42	169.62	1.98
Lahma Reservoir [77]	177	131	16.28	238	1.69
Ghezala Reservoir [29]	61	118	27.42	204.49	1.5
Tall Lake (Philippines) [78]	71–76	150–180	14–17	250–300	1.57
China [79]	100–150	120–150	>50	600–800	4.625–5.06
Southern United States [80]	50	112	30	250	1.96

Table 10.
Comparative growth performance of Nile Tilapia in different environments.

pens placed in Smati Reservoir (ICJ = 0.4–1.54 g/d) against 0.65 g/d in the cages of Bechima Station.

On the other hand, we recorded very important survival rates compared to those obtained by other authors. This shows that Tunisian fresh waters and geothermal waters allow the growth and survival of this species. However, we recommend the maintenance of favorable conditions for the growth and survival of individuals including fish sorting which must be conducted in the future into 3 enclosures and 3 cages in order to homogenize the stock.

To highlight the results found in this study, we compared them with other authors' findings (**Table 10**).

The results show that the growth values found during this study are slightly below those attained by other researchers. On the other hand, fish grown in freshwater show daily growth rates close to those estimated by other authors who have worked in Tunisian reservoirs. In China, reported growth rates are high due to the use of large fry at the beginning of the grow-out phase and the good quality of the supplemented feed.

7. Conclusion

The present work contributes to the literature on Nile Tilapia farming "*O. niloticus*" in fresh waters (using enclosures placed in Smati Reservoir) and in geothermal waters (deploying cages installed in the large basin of Bechima experimental station) in Tunisia. This study was made March and August 2017, a period with abiotic conditions which are optimal for the rearing of this species. Larval rearing monitoring in the nursery located at the level of the greenhouse in Bechima Station has shown that the earliest larval stages are much more vulnerable than the most advanced stages. This vulnerability can be explained by the physical and physiological weakness associated to the fragility of larvae during their early stages of development, as well as their sensitivity to the manipulations associated to the rearing procedure. The larvae used in this study were collected from 13 broodstock in March 2017. During the 60 days of larval rearing, the average final weight increased from 0.01 to 0.85–2.55 g, and the survival rate recorded (70%) was within the range of values found by other authors.

The analysis of the evolution of the physico-chemical parameters measured in situ (temperature, dissolved oxygen, salinity and pH) as well as nutrients (nitrates, nitrites, ammonium and orthophosphates) shows that the values are well within the range recommended for Nile Tilapia rearing.

The preliminary results of the breeding phase in cages placed in the grow-out basin in Bechima station and in pens set in Smati Reservoir are quite encouraging. The comparison of the results obtained in cages placed in geothermal waters and in pens placed in the reservoir, has demonstrated that the breeding of frays in fresh waters shows a much better growth performance than in geothermal waters. The conversion rate obtained in fry, with body weights ranging from 3 to 19 g, reared in pens (0.87) is lower than that obtained in cages (1.62 and 1.58). In the grow-out phase, the results obtained after 30 days show that the fish reared in freshwater maintain the best growth performance compared to that reared in geothermal water. The preliminary results of the grow-out phase in Smati Reservoir are very encouraging compared to those achieved in Bechima Station. This observation highlights the great potential of this species' breeding and seeding in Tunisian reservoirs. The mortalities recorded in the two study areas are essentially due to cannibalism associated with the heterogeneity of the sizes and masses of the reared fish.

The in-depth diagnosis of this activity as well as the global evaluation of the results reveal the presence of anomalies and problems that must be addressed for the next reproduction and larval rearing campaigns in order to better manage this process and improve the production in the hatchery.

The results obtained shows that this type of fish farming has significant production potential. However, it presents two problems that must be solved, namely the massive production of calibrated fry of *O. niloticus* and the elaboration of a quality feed to meet the nutritional needs of fish. Thus, important improvements remain to be made in feeding and nutrition in order to specify and determine the standards of manufacturing and distribution of fish feed. This would ensure increased production and economic profitability along with a better coverage of the feed requirements of tilapia taking into account the economic context and the locally available compounds.

Acknowledgements

The authors extend their gratitude to the IRESA which provided financial support for the impact research project (AMBISEPT) from which stems this research paper.

They are similarly thankful to all the staff of the Technical Center of Aquaculture (Tunisia), the General Directorate of Fisheries and Aquaculture (Tunisia) as well as the students of HIFA OF Bizerte for the efforts they exerted during the realization of the practical part of this work.

Conflict of interest

The author declares no conflict of interest.

Author details

Sami Mili[1]*, Rym Ennouri[1], Manel Fatnassi[1], Hajer Zarrouk[1], Rabeb Thabet[1] and Houcine Laouar[2]

1 University of Carthage, Higher Institute of Fishery and Aquaculture of Bizerte, Research Unit: Exploitation of Aquatic Environments, Bizerte, Tunisia

2 Technical Center of Aquaculture, Tunis, Tunisia

*Address all correspondence to: mili.sami.ispa@gmail.com

References

[1] FAO. FAO Fisheries & Aquaculture - Cultured aquatic species fact sheets - *Oreochromis niloticus* (Linnaeus, 1758) [Internet]. 2017. Available from: http://www.fao.org/fishery/culturedspecies/Oreochromis_niloticus/fr [Accessed: August 12, 2017]

[2] Lazard J, Baruthio A, Mathé S, Rey-Valette H, Chia E, Aubin J, et al. Adaptation des typologies d'exploitations aquacoles aux exigences du développement durable. Cahiers Agricultures, EDP Sciences. 2009; **18**(2–3):199-210. DOI: ff10.1684/agr.2009.0297ff.ffhal-01460889f

[3] Trinh TQ, Agyakwah SK, Khaw HL, Benzie JAH, Attipoe FKY. Performance evaluation of Nile tilapia (*Oreochromis niloticus*) improved strains in Ghana. Aquaculture. 2021;**2021**(530):735938. DOI: 10.1016/j.aquaculture.2020.735938

[4] FAO. Nicaragua, Fisheries and Aquaculture Division [Internet]. 2022. Available from: https://www.fao.org/fishery-aquaculture/en [Accessed: April 3, 2022].

[5] Philippart JC, Ruwet JC. Chapter: Ecology and distribution of tilapias. In: Pullin, Lowe-McConel, editors. The Biology and Culture of Tilapias. Manila, Philippines: ICLARM Conference Proceedings, Int. Center for Living Aquatic Resources Management; 1982. pp. 15-59

[6] Assane Anabi T, Issiaka Y. Diversité ichtyologique d'un petit lac tropical: cas du lac Madarounfa, Niger. Revue Ivoirienne des Sciences et Technologie. 2021;**37**:143-154

[7] Azaza MS. Optimisation de l'élevage intensif du Tilapia de Nil *Oreochromis niloticus* (L., 1785), dans les eaux géothermales de Sud Tunisien : Effet de l'alimentation et de la température sur les performances de croissance [Thèse]. Université de Tunis Faculté des Sciences de Tunis; 2009

[8] Dhraief M, Azaza MS, Kraiem M. Étude de la reproduction du Tilapia du Nil *Oreochromis niloticus* (L.) en captivité dans les eaux géothermales du sud tunisien. Bulletin de l'Institut National des Sciences et Technologies de la Mer (INSTM). 2010;**37**:89-96

[9] FAO. Fisheries & Aquaculture - Cultured aquatic species fact sheets - *Oreochromis niloticus* (Linnaeus, 1758) [Internet]. 2005. Available from: http://www.fao.org/fishery/culturedspecies/Oreochromis_niloticus/fr [Accessed: August 8, 2017]

[10] Mary A. Infestation of Ectoparasites on Nile Tilapia (*Oreochromis niloticus*) in Aquaculture Production in the Ashanti Region, Ghana [Thesis]. Norway: Department of Marine and Freshwater Biology, Norwegian College of Fishery Science and University of Tromsø; 2006

[11] Azaza MS, Mensi F, Abdelwaheb A, Kraiem M. Elaboration d'aliments secs pour Tilapia du Nil *Oreochromis niloticus* (L., 1758) en élevage dans les eaux géothermales du sud tunisien. Bulletin de l'Institut National des Sciences et Technologies de la Mer (INSTM). 2005;**32**: 23-30

[12] Azaza MS, Wassim K, Mensi F, Abdelmouleh A, Brini B, Kraïem MM. Evaluation of faba beans (*Vicia faba L. var. minuta*) as a replacement for soybean meal in practical diets of juvenile Nile tilapia *Oreochromis niloticus*. Aquaculture. 2009;**287**:174-179. DOI: 10.1016/j.aquaculture.2008.10.007

[13] Thabet R. Etude comparative de l'élevage du Tilapia du Nil "*Oreochromis niloticus*" entre les eaux douces et géothermales en Tunisie [Master]. Tunisie: Institut Supérieur de Pêche et d'Aquaculture de Bizerte et Centre Technique d'Aquaculture; 2017

[14] Zaouali J. Problèmes d'aquaculture : eaux saumâtres et potentiel aquacole. Archives de l'Institut Pasteur de Tunis. 1981;**58**(1–2):93-103

[15] Dhraief M. Reproduction du Tilapia du Nil *Oreochromis niloticus* (L., 1758) [Teleostei, Cichlidae], en captivité dans les eaux géothermales du sud Tunisien: Effet de quelques facteurs démographiques et environnementaux [Master]. Tunisie: Institut Supérieur de Pêche et d'Aquaculture de Bizerte; 2005

[16] Azaza MS. Tolérance à la température et à la salinité chez le tilapia du Nil (*Oreochromis niloticus* L., 1758) en élevage dans les eaux géothermales du sud tunisien [Master]. Tunisia: Faculty of Sciences of Tunis; 2004

[17] Derouiche E, Azaza MS, Kraiem M. Essai d'acclimatation du Tilapia du Nil, *Oreochromis niloticus* dans la retenue de barrage de Lebna (Cap bon, Tunisie). Bulletin de l'Institut national des sciences et technologies de la Mer. 2009; **36**:97-92

[18] DGEQV. Aquaculture en Tunisie: Impacts environnementaux et potentialités de développement. Phase I: Tunisia: Etat du secteur aquacole en Tunisie. Direction Générale de l'Environnement et de la Qualité de la Vie; 2008. p. 246-308

[19] Rhouma A, Ouaer E. L'aquaculture en Tunisie: Potentialités et techniques d'élevage. Bulletin de l'Institut National des Sciences et Technologies de la Mer (INSTM). 1978;**4**(78):43

[20] DGPA (Direction Générale de la pêche et de l'aquaculture). Annulaire des Statistiques des pêches et aquaculture en Tunisie, Ministère de l'agriculture [Internet]. 2016. Available from: http://www.ispab.agrinet.tn/docs/annuaire/stat2016.pdf [Accessed: May 1, 2021]

[21] DGBGTH. Catalogue des documents, Projet GE.O.R.E. Ministère de l'Agriculture et des Ressources Hydrauliques/Coopération Technique Tuniso-Allemande/GTZ; Tunisia. 2008. p. 51

[22] FAO. Tilapia du Nil [Internet]. 2017. Available from: http://www.fao.org/fishery/affris/profil-des-especes/nile-tilapia/tilapia-du-nil-accueil/fr/ [Accessed: June 6, 2022]

[23] FAO. La situation mondiale des pêches et de l'aquaculture 2018. Atteindre les objectifs de développement durable. Rome: Organisation des Nations Unies pour l'Alimentation et l'Agriculture; 2018. p. 237

[24] Trewavas E. Tilapiine Fishes of the Genera *Sarotherodon Oreochromis* and Danakilia. London: British Museum (Natural History); 1983. p. 583

[25] Santos VB, Mareco EA, Silva MDP. Growth curves of Nile tilapia (*Oreochromis niloticus*) strains cultivated at different temperatures. Acta Scientiarum Animal Sciences. 2013;**35**:235-242. DOI: 10.4025/actascianimsci. v35i3.19443

[26] Robisalmi A, Alipin K, Gunadi B. Effect of different feed restrictions on growth, biometric, and hematological response of juvenile Red Tilapia (*Oreochromis spp*). Jurnal Ilmiah Perikanan dan Kelautan. 2021;**13**(2):243. DOI: 10.20473/jipk.v13i2.26302

[27] Rakotomalala T. Faisabilité technique et économique des mises en

place d'un élevage en cage de *Tilapia nilotica* au lac Kavitaha (AMPEFY). Antananarivo, Madagascar: ESSA (Etablissement Supérieur des Sciences Agronomiques); 2004. p. 91

[28] Adjanke A. Production d'alevins et gestion de ferme piscicole. Coordination togolaise des organisations paysanes et de producteurs agricoles. Lom, Bulgaria: C.T.O.P; 2011. p. 39

[29] CTA. Centre Technique de l'Aquaculture: Grossissement du Tilapia du Nil dans les eaux géothermales. Tunis: Rapport du ministère de l'agriculture et de l'environnement de Tunisie; 2012. p. 10

[30] CTA. Echos de l'aquaculture: Optimisation de la production d'alevins de Tilapia du Nil « *Oreochromis niloticus* » dans la station de Boumhel. Vol. 4. Centre Technique de l'Aquaculture; 2015. p. 24

[31] Tilapia SV. In: Lucas JS, Southgate PC, editors. Aquaculture: Farming Aquatic Animals and Plants. NJ, USA: Blackwell Publishing; 2003. pp. 312-345

[32] CTA. Centre Technique de l'Aquaculture. Fiche espèce: Le Tilapia du Nil *Oreochromis niloticus*. Tunis: Fiche de l'Aquaculture continentale en Tunisie; 2017. p. 2

[33] Mélard C. Les bases biologiques de l'élevage du Tilapia du Nil. Cahiers d'Ethologie Appliquée. 1986;**6**(3):224

[34] Sifa L, Chenhong L, Dey M, Gagalac F, Dunham R. Cold tolerance of three strains of Nile tilapia, *Oreochromis niloticus*. Aquaculture. 2002;**2002**(213): 123-129. DOI: 10.1016/S0044-8486(02) 00068-6

[35] Chervinski J. Environmental physiology of tilapias. In: Pullin RSV, Lowe-McConnell RH, editors. The Biology and Culture of Tilapia. Manila, Philippines: ICLARM Conference Proceedings; 1982. pp. 119-128

[36] Kayansamruaj P, Pirarat N, Hirono I, Rodkhum C. Increasing of temperature induces pathogenicity of *Streptococcus agalactiae* and the up-regulation of inflammatory related genes in infected Nile tilapia (*Oreochromis niloticus*). Veterinary Microbiology. 2014;**172**(1): 265-271. DOI: 10.1016/j.vetmic.2014. 04.013

[37] Marcusso PF, Aguinaga JY, Claudiano GS, Eto SF, Fernandes DC, Mello H, et al. Influence of temperature on *Streptococcus agalactiae* infection in Nile tilapia. Brazilian Journal of Veterinary Research and Animal Science. 2015;**52**(1):57-62. DOI: 10.11606/ issn.1678-4456.v52i1p57-62

[38] Lazard J. Les systèmes aquacoles face au changement climatique. Cahiers Agricultures. 2017;**26**(3):34001. DOI: 10.1051/cagri/2017018

[39] Huet M. Traité de pisciculture. Bruxelles: Éditions Ch. de Wyngaert; 1970. p. 718

[40] Benidiri R. Création d'un projet piscicole [Master]. Tlemcen-Algeria: Faculté de Technologie, Département de Genie Electrique et Electronique; 2017

[41] El Asely AM, Reda RM, Salah AS, Mahmoud MA, Dawood MAO. Overall performances of Nile tilapia (*Oreochromis niloticus*) associated with using vegetable oil sources under suboptimal temperature. Aquaculture Nutrition. 2020;**26**(4):1154-1163. DOI: 10.1111/anu.13072Citations: 19

[42] Azaza MS, Mensi F, Kraiem MM. Grossissement du Tilapia du Nil (*Oreochromis niloticus*, L., 1758): en

bassins dans les eaux géothermales du sud tunisien et en cages flottantes dans la retenue du barrage de sidi saâd (centre de la Tunisie). Communication à la 8ème Conférence Internationale des Limnologues d'Expression Française (CILEF 2006). 2006. Available from: https://www.researchgate.net/publication/260262892_Grossissement_du_Tilapia_du_Nil_Oreochromis_niloticus_L_1758_en_bassins_dans_les_eaux_geothermales_du_sud_tunisien_et_en_cages_flottantes_dans_la_retenue_du_barrage_de_sidi_saad_centre_de_la_Tunisie_C [Accessed: Tuesday, 2023-01-25].

[43] Kraiem M, Azaza MS. Etude de la tolérance a la température et à la salinité chez le tilapia du Nil *Oreochromis niloticus* (L.) élevé dans les eaux géothermales du sud tunisien. Bulletin de l'Institut National des Sciences et Technologies de la Mer (INSTM). 2007;**34**(2):145-155

[44] Stickney R. Tilapia tolerance of saline water. Review. The Progressive Fish-Culturist. 1986;**48**:161-167

[45] Song L, Zhao Y, Song Y, Zhao L, Ma C, Zhao J. Effects of saline-alkaline water on growth performance, nutritional processing, and immunity in Nile tilapia (*Oreochromis niloticus*). Aquaculture. 2021;**544**(15):737036. DOI: 10.1016/j.aquaculture.2021.737036

[46] Ait-Hamouda I. Contribution à l'étude de l'inversion sexuelle chez une espèce de poisson d'eau douce: Tilapia (*Oreochromis niloticus*) [Mémoire ingénieur]. Aquaculture. Ismal-Algeria: Inst. Science de la mer; 2005

[47] Balarin J, Hatton J. Tilapia. A Guide to their Biology and Culture in Africa. Stirling (UK): Univ of Stirling; 1979. p. 203

[48] Leprieur F, Rubin A. INVAQUA: Prédiction de l'établissement des espèces exotiques dans les milieux aquatiques : vers une anticipation des invasions biologiques. Convention Onema-MNHN 2010 Dynamique des biocénoses aquatiques (Action 7). Paris. 2011:131

[49] Badiane I. Effets de la substitution de la farine de poisson par la poudre de feuilles de *Moringa oleifera* (Lam.) sur la croissance des juvéniles du tilapia du Nil (*Oreochromis niloticus*) (Linnaeus, 1758) [Master]. Senegal: University of Gaston Berger; 2019

[50] Rappaport A, Sarig S, Marek M. Results of tests of varius aeration systems on the oxygen regime in the Genosar experimental ponds and growth of fish there in 1975. Bamidgeh. 1976;**28**(3):35-49

[51] Benabdellah N. Etude expérimentale sur l'activité des enzymes digestives (trypsine et chymotrypsine) chez les alevins du tilapia du Nil (*Oreochromis niloticus*) (Linnaeus, 1758) en relation avec la qualité du régime alimentaire protéique distribué [Master]. Algeria: Oran University; 2011

[52] Adoumandjali G. Variabilité génétique, moléculaire et quantitative du Tilapia du Nil (*Oreochromis niloticus*, Linnaeus, 1758) dans le bassin du Congo [Thèse]. Québec, Canada: Université LAVAL; 2020, 2020

[53] Malcolm C, Beverdije H, McAndrew B. Tilapias: Biologie and Exploitation. Institute of Aquaculture. University of Stirling, Scotland. Kluwer Academic Publishers; 2000. p. 185

[54] Mélard C. Les bases biologiques de l'élevage du Tilapia du Nil. Belgium: Institute of Zoology, University of Liège; 1989. p. 224

[55] El Sayed AF, Mamdouh K. Effects of photoperiod on the performance of

farmed Nile tilapia *Oreochromis niloticus*: I. growth, feed utilization efficiency and survival of fry and fingerlings. Elsevier Science. 2004;**231**(1–4):393-402. DOI: 10.1016/j.aquaculture.2003.11.012

[56] FAO. *Oreochromis niloticus*. Cultured Aquatic Species Information Programme: Division des pêches et de l'aquaculture [Internet]. 2022. Available from: https://www.fao.org/fishery/fr/culturedspecies/oreochromis_niloticus_fr/fr [Accessed: May 5, 2022].

[57] Kestemont P, Micha J, Falter U. Les méthodes de production d'alevins de Tilapia Nilotica, programme de mise en valeur et la coordination de l'aquaculture. ADCP/REP/89/46; 1989. 132.

[58] Azaza MS, Legendre M, Kraiem M, Baras E. Size-dependent effects of daily thermal fluctuations on the growth and size heterogeneity of Nile tilapia *Oreochromis niloticus*. Journal of Fish Biology. 2010;**76**(3):669-683

[59] Lazard J, Legendre M. La reproduction spontanée du tilapia: une chance ou un handicap pour le développement de l'aquaculture africaine. In: RSV P, Lazard J, Legendre M, JB AK, Pauly D, editors. Le Troisième Symposium International sur le Tilapia en Aquaculture. Montpellier, France: ICLARM. Conf. Proe; 1996. pp. 82-98

[60] Mélard C, Philippart J. La production de tilapia de consommation dans les rejets industriels d'eau chaude en Belgique. Cahiers d'Ethologie appliquée, collection Enquêtes et dossiers: 2. 1981;1(suppl. 2): 122.

[61] Little D, Bhujel R, Pham T. Advanced nursing of mixed-sex and mono-sex tilapia (*Oreochromis nilotcus*) fry, and its impact on subsequent growth in fertilized ponds. Aquaculture. 2003; **221**:265-276

[62] Campbell D. Formulation des aliments destinés à l'élevage de *Tilapia Nilotica* (L.) En cages dans le lac de Kossou Cote D'Ivoire. Rapport Technique, Authorite Aménagement Vallée du Bnadama. 1978;**45**:31

[63] Mili S, Ennouri R, Fatnassi M, Laouar H, Zarrouk H, Chargui1 T. Technical notes on structures of small-scale freshwater fish farming in Tunisia. Journal of Aquaculture & Marine Biology. 2021;**10**(5):218-223. DOI: 10.15406/jamb.2021.10.00324

[64] Turki I, Kraiem M. Séminaire international sur l'aquaculture en Tunisie. In: Pisciculture d'eau douce; Mars 2002. Tunisie: APIA Tunis; 2002. p. 12

[65] Le Cren E. The length-weight relation-ship and seasonal cycle in gonad weight and condition of the perch (*Perca fluviatilis*). The Journal of Animal Ecology. 1951;**20**:210-219

[66] Mili S, Jarboui O, Missaoui H. Caracteres biometriques de la squille Squilla mantis dans les eaux tunisiennes. Bulletin de l'Institut National des Sciences et Technologies de la mer. 2008;**35**:1-14

[67] NSTM. Résultats des recherches valorisables en aquaculture: Optimisation de la production d'alevins de Tilapia du Nil «*Oreochromis niloticus*» dans la station de Boumhel. Echos de l'aquacult. 2009. p. 24

[68] Coulibaly N. Relation taille-poids de 11 espèces de poissons du Burkina Faso. In: Palomares MLD, Samb B, Diouf T, Vikily JM, Pauly D, editors. Fish Biodiversity: Local Studies as Basis for Global Interferences. 2003;**14**:281

[69] Du Feu N. Population parameters for six commercial species in Lake Kainji, Nigeria, using lenght frequency data sampled from artisanal fish catches. In: Palomares MLD, Samb B, Diouf T, Vikily JM, Pauly D, editors. Fish Biodiversity: Local Studies as Basis for Global Interferences. 2003. p. 292

[70] Babiker M, Ibrahim H. Studies on the biology of reproduction in the cichlid *Tilapia nilotica* (L.): Gonadal and fecundity. Journal of Fish Biology. 1979; **14**:437-448

[71] Coward K, Bromage N. Spawning periodicity, fecundity and egg size in laboratory-held stocks of a substrate-spawning tilapiine, *Tilapia zillii* (Gervais). Aquaculture. 1999;**171**(3–4): 251-267. DOI: 10.1016/S0044-8486(98) 00498-0

[72] Philippart JC, Ruwet J. Ecology and distribution of tilapias. In: Pullin RSV, LoweMcConnell RH, editors. The Biology and Culture of Tilapia: ICLARM Conférence Proceedings. 1982. pp. 15-59

[73] FAO. The Stats of World Fishers and Aquaculture. Rome: Food and Agriculture Organization of the United Nation; 2012. p. 230

[74] Legendre M. Potentialités aquacoles des Cichlidae (*Sarotherodon melanotheron*, Tilapia guineensis) et Clariidae (*Heterobranchus longifilis*) autochtones des lagunes ivoiriennes [Thèse]. Université de Montpellier II; 1991

[75] Eer A, Schie T, Hilbrands A. La pisciculture en eau douce à petite échelle. Agromisa. 2004:85

[76] Mehak A, Mu Y, Mohsin M, Noman M, Nazir K. Population dynamics of Nile Tilapia (*Oreochromis niloticus*) at Chashma Barrage, Pakistan. Indian Journal of Geo-Marine Sciences. 2017;**46** (01):206-210

[77] Laouar H, Barhoumi L. Approche technico-économique de l'élevage en cages de Tilapia du Nil dans les retenues de barrages tunisiennes 2017. p. 9

[78] Bestari N, Edwards P, Katon B, Morales A, Pullin R. An evaluation of small scale freshwater rural aquaculture development for poverty reduction. Case study 6: Tilapia Cage Farming in Lake Taal, Batangas. Philippines Report: Asian Development Bank. 2005;**091704**: 110-127. ISBN 971-561-550-3

[79] Qiuming L, Yang Y. Tilapia Culture in Mainland China (Paper Presented at the Sixth International Symposium on Tilapia in Aquaculture, 12–16 September 2004). Vol. 2004. Philippine International Convention Center; 2004. pp. 18-27

[80] McGinty AS, Rakocy JE. Cage Culture of Tilapia. Southern Regional Aquaculture Centre. US: SRAC Publication; 1999. p. 281

Section 3

Innovation and Technology

Chapter 6

Perspective Chapter: Alternative Intensive Animal Farming Tactics That Minimize Negative Animal Impact and Improve Profitability

Songul Senturklu, Douglas Landblom, Gerald Stokka and Larry Cihacek

Abstract

Animal agricultural businesses strive to improve efficiencies, reduce input costs, and maintain healthy animals with minimal disease control intervention. Bovine respiratory disease is a disease complex that increases when cattle are reared in confinement costing the North American beef cattle industry three-billion dollars or more annually. Principles of soil health define the need to reduce tillage, keep the soil surface covered, rotate crops and plant cover crops for greater plant diversity, maintain living roots in the soil for as long as possible, and integrate livestock grazing into cropping systems. As beef calves age they experience more viral and microbial challenges which stimulate an immune system response resulting in greater disease resilience and well-being when commingled with unfamiliar cattle for confinement feedlot finishing. Wintering calves after weaning in November for modest growth of 0.59 kg/day (1.30 lbs./day) combined with integrated grazing of a sequence of native range and annual forages grown in a diverse multi-crop rotation is a management mechanism that increases calf age (200^+ days), promotes structural growth, and delays feedlot entry. Retaining ownership using a vertically integrated business model from birth to slaughter accounting for all business inputs and outputs has resulted in improved environmental balance and business profitability.

Keywords: beef cattle, bovine respiratory disease, sequential grazing, reduced concentrated feeding, integrated crop-livestock system, regenerative agriculture, animal welfare, reduced production cost, net return, profitability

1. Introduction

In animal agriculture, well managed businesses are structured around important intricacies that maintain and improve efficiencies, and managers of beef cattle enterprises continually strive to reduce production cost. The cow-calf business generates calves from which the entire remaining cattle industry relies upon and as such initial management weighs heavily on how feeder cattle perform from weaning to final harvest. Sound management is key to animal wellbeing and the methods used to minimize stress. Bovine respiratory disease (BRD) in North America is the most studied beef cattle illness costing the beef cattle industry in excess of three billion dollars annually due to reduced animal performance and depressed carcass quality, death loss, pharmaceuticals, and expenses related to treatment [1]. Due to stressors such as commingling, transportation and dehydration a number of calves will experience BRD after feedlot arrival. To reduce the risk of early onset BRD, feedlot operators will administer antimicrobials to animals during processing. This process known as metaphylaxis or prophylactic use is applied to all animals in the high-risk group [2]. Abrupt weaning is a stressful event, when coupled with commingling on the ranch of origin adds further stress that increases the risk of BRD. Therefore, stepwise pre-weaning management strategies to reduce stress prior to weaning that includes specific vaccination protocols, and commingling prior to weaning will reduce the impact of separating calves from their dams. In addition to calf hood vaccination protocols at one to two months of age, a 42-day pre-weaning vaccination preconditioning program beginning six and three (booster) weeks before weaning that includes introduction to dry feed (self-fed creep feed supplement) reduces stress and results in a greater number of animals developing a protective immune response before weaning. An extensive BRD metanalysis review conducted by Taylor et al. [3] revealed inconsistencies that made across study evaluations difficult when evaluating processing methods, vaccination, preconditioning, nutritional factors, and prophylactic methods that include administration of antimicrobial metaphylaxis. The authors concluded that BRD can be best managed using preconditioning techniques coupled with weaning before selling and that calf age is important. Calves that experience more viral and microbial challenges develop immune defenses with aging and are more resilient to viral and microbial insults after feedlot arrival.

The beef cattle industry is segmented into geographical regions based on available feed, water, labor supply, environment, and ease of ground transport. Confinement cattle feeding businesses cannot operate without a supply of feeder cattle from cow-calf producers. For the most part, cows that produce a supply of calves for the cattle feeding industry are managed in geographical regions unsuitable for crop production and from mixed crop and livestock farms. As such, cow-calf producers selling calves as the first point of sale is the time that new wealth is generated. All future purchases and sales are based on a buy/sell margin plus interest expense culminating in either profit or loss. Based on market conditions and the potential for future profit (loss), cow-calf producers can decide to sell 6–8-month-old calves after weaning and repeat the cycle annually, or retain ownership through a growing period (backgrounding) and sell their calves after approximately 100 days on feed. Thus, producers are subject to a series of keep/sell marketing management decisions occurring from weaning through finishing and final harvest. Keep/sell decisions parallel seasonal management and availability of sufficient winter feed supply, spring-summer-fall pasture, and market projections that either do, or do not, support retained ownership. With respect to BRD morbidity and mortality, weaning prior to sale is the largest contributor to the reduction in BRD morbidity. Therefore, when calves have been weaned, processed (viral and clostridial pathogen vaccinations, castration, and dehorned) and fed post-weaning diets for at least six weeks (preconditioned), the incidence of BRD morbidity and mortality is reduced 4.5 times, but not eliminated [4].

Retaining ownership in a vertically integrated business model from birth to final harvest has been shown to result in enhanced compensatory gain and efficiency, reduced days on feed and breakeven expense, and profitability increase, when feedlot entry was delayed until after extended grazing of forages [5–7]. Research involving the integration of a diverse multi-crop farming system coupled with beef cattle grazing was designed to evaluate the impact of combining the two enterprises on extensive rearing and animal health, soil health, crop production, grazing animal performance and economics, and the effect of delayed feedlot entry on system profitability.

2. Beef cattle management in the United States

To better understand the manner in which alternative intensive animal farming tactics can minimize negative effects on animal health, it is important to understand the multiple ways beef cattle and calves are managed and marketed after weaning.

Initially, beef cattle producers are faced with the marketing decision of when to sell calves. The decision comes down to whether calves will be sold immediately after weaning, preconditioned for a period of 42-days before selling, or ownership retained for a longer period of time. Before the backgrounding period begins, the producer must determine that there is sufficient feed available to feed the calves and when the projected backgrounding period will end. The next questions the producer must answer are: 1) do I sell the calves at the end of the backgrounding period, or 2) keep the calves and put them on spring and summer pasture for summer grazing. The market timing decision is important, because dietary energy level during the growing period effects future performance, especially cattle destined for summer grazing. Steers and heifers destined to go to the feedlot after the growing period can be fed dietary energy levels that support average daily gain (ADG) of 1.14 to 1.36 kg/hd/day (2.5 to 3.0 lb/hd/day). However, if steers and/or heifers are destined for summer grazing of perennial and annual forages, then (ADG) of 0.59 kg/hd/day (1.30 lb/hd/day) is a more appropriate confinement pre-grazing growth rate, because although early spring vegetative pasture grasses are highly nutritious, high water content ($\geq$80%) of early spring vegetative grasses and the quantity of available forage can restrict the animals' ability to consume a sufficient quantity of dry matter for maintenance and growth [8–10]. Steers and heifers fed higher energy diets for more rapid growth during the drylot wintering period, in the Northern Great Plains, will have greater body fat that cannot be maintained when transitioned to a grazing environment. Therefore, a high energy pre-grazing dietary regimen is inappropriate for animals destined for a grazing environment, because body condition will decline until a grazing growth equilibrium is attained. The amount of time for equilibrium to occur depends on the degree of fatness and forage quality. In addition to appropriate pre-grazing body condition, beef cattle heifers placed on grass that are not intended for breeding purposes will experience reduced ADG due to physical activity associated with estrus activity. Estrus activity is easily alleviated with ovariectomy conducted by a licensed veterinarian. At the end of the summer/fall grazing period, the producer determines whether to continue grazing late into the fall period, sell, or retain ownership and place the yearlings in the feedlot. Grazing late into the fall and early winter period in the Northern Great Plains region of the United States results in slower than desired ADG due to declining forage quality. When retaining ownership late into the fall/early winter period, an alternative to grazing low quality pastures is to feed harvested round baled hay in a free-ranging environment using a technique referred to as "bale grazing". In this situation, the animals are not confined to feedlot pens, but are allowed to range freely while being fed high quality baled hay. For intensified animal agriculture, feeding areas might range from 65.0 to 135.0 ha in size; however, the feeding areas can be sized to fit a given number of feeder cattle. Upon completion of the bale grazing period, the producer decides whether to continue retaining ownership by placing the feeder cattle in a feedlot or to sell. Assuming the decision is to retain ownership, another decision needs to be made that will affect how the animals are to be fed. That decision is whether the cattle will be forage finished using a forage-based diet and non-starch finishing supplement [11] or fed a high-caloric grain-based feedlot energy diet designed for 2.15 to 2.27 kg/hd/day (4.75 to 5.0 lb/hd/day).

3. Climate, crops and regenerative agriculture, diversified cropping system

3.1 Climate

The semi-arid region of the Northern Great Plains is known for wind, cold winters, and warm summers; however, relatively low humidity. Growing season (April – October) precipitation averages 311.9 mm. Maximum and minimum mean temperatures range from a high of 23.8 °C to a low of 8.5 °C [12].

3.2 Crops and regenerative agriculture

A wide range of crops are grown in the region for grain, oilseed, and forage including numerous varieties of, but are not limited to: cereal grains (spring wheat, winter wheat, corn, barley, oats, rye, flax, triticale, lentil, chickpea, grain sorghum, dry beans, dry pea), oilseeds (soybean, sunflower, canola, safflower, crambe), and forages (alfalfa, clover, millet, hairy vetch).

Alternative approaches to minimizing animal health issues focuses on methods whereby cattle are managed to spend upwards of 85% of their lives outside of feedlot confinement. Initially, pasture stocking rate for cows and calves that will be grazing a given range resource is determined. Secondly, the number of cow-calf stocking spaces that retained ownership yearlings will replace needs to be determined. For crop and livestock farms, an integrated diversified multi-crop rotation system can provide additional grazing without a large reduction in the ranch's cow-calf carrying capacity.

When designing the annual cropping system, complementary attributes were considered from the perspective of the following: minimum soil disturbance using no-till seeding and planting, suitability for cattle grazing, water conservation, crops that form associations with arbuscular mycorrhizal fungi (AMF), improving soil quality through soil nutrient cycling, including legume crops or mixes with a high percentage of legumes in the mix, crops that have high root mass, deep cycling crops, and crops that maintain a living root in the soil long after freezing conditions set in. More recently, coalescing these non-traditional practices and applying them to farming and ranching has become referred to as regenerative agriculture. This is not a term often heard around traditional farming circles. However, among holistic farmers and ranchers, regenerative agricultural practices focus on melding this wide range of practices together in ways that are good for the land and the people who farm the land. Soil is a living organism and must be managed carefully, because soil coupled with water, solar radiation, and microbial derived nutrient cycling sustains all plants and living creatures. Regenerative agriculture has a foundation in the five principles of soil health: Soil Armor, Minimizing Soil Disturbance, Plant Diversity, Maintaining a Continual Live Root in the Soil as Long as Possible, and Livestock Integration [13].

The soil surface is fragile and subject to wind and water erosion as well as impact from insults such as hail and solar heat that kills soil surface microbes. Protection for soils comes from plant cover of pastures, farmed land with domestic no-till or reduced-till crop production and residues, cover crop mixes that help keep the land covered and provide forage for haying and grazing, and reduced weed infestation.

3.3 Diversified cropping system

Considering the wide array of crops that could be grown and demonstrate complementarity, the diversified multi-crop rotation consisted of spring wheat, dual winter and summer cover crop, forage corn, field pea-forage barley mix, and sunflower. Within these crops, cool- and warm-season grass and broadleaf crops are represented that are adapted to the semi-arid region. Crop characteristics associated with crops selected for inclusion in the diverse crop rotation are shown in **Table 1**. The characteristics listed include Crop Type (Cool- or Warm-season Grass and Broadleaf types), crop water use requirement (Low, Medium, High), Grain Crude Protein %, Residue C:N Ratio, Nitrogen Scavenging Ability, and whether the crop forms an association with Arbuscular Mycorrhizal Fungi. Some excellent crops were excluded that did not meet the requirement for livestock grazing or did not form relationships with AMF. For example, canola is an excellent oilseed crop; however, the crop is not suitable for grazing and does not form a relationship with AMF. Nonetheless, within a wider rotation of six to seven crops canola and soybean would be logical crop rotation additions.

Cover crops were initially promoted by USDA/NRCS for purpose of controlling water and wind erosion prescribed by Practice Code: 340: Growing a crop of grass, small grain or legumes primarily for seasonal protection and soil improvement [14]. Ancient alternatives to fertilizers were the use of green manure crops that were used by farmers in Chinese, Greek, and Roman societies [15]. In the infancy of cover crop use the recommendation was that the crop would be seeded following a primary cash

Crop	Spring wheat	Multi-specie cover crop	Corn	Field pea/ barley mix	Sunflower
Crop Type	Cool-Grass	All Crop Types	Warm-Grass	Cool- Grass & Broadleaf	Warm-Broadleaf
Water Use	Medium	Medium	High	Low/Low	High
Crude Protein % (Grain)	12-16	Mixed	8-9	24/13	20-28
Crop Residue C:N Ratio	90	30-45	57	27/80	68
N Scavenging Ability	Very Good	Very Good Varies with mix	Deep soil mining	Fixation/ Very Good	Deep soil profile mining
Forms Arbuscular Mycorrhizal Fungi Association	Yes-Medium	Yes-High	Yes-High	Yes-Medium	Yes-Medium

Table 1.
Multi-crop rotation crop characteristics.

crop, which has not been very successful in the semi-arid regions. This is because dry soil conditions are common after a primary crop is harvested and germination is impeded reducing biomass production. Mixed-specie cover crops are gaining popularity among crop and livestock producers for their aid in not only controlling wind and water erosion, but also as full-season annual forage crops used for haying or grazing that provide for both above and below ground biomass. Cover crops provide numerous soil system services beyond protecting the soil surface from erosion. Soil system services also include increasing fertility from soil organic matter and subsequent nitrogen supplied from decaying roots and surface residue, symbiotic and asymbiotic nitrogen fixation, increased soil aggregation and decreased compaction, increased soil water infiltration through the use of tillage-type radish and turnips that create tunnels for soil water infiltration, weed control to some extent, grazing season extension, and protective cover for wildlife. To accomplish these many soil system services within the research investigation, the diverse crop rotation employed a dual winter and summer cover crop planting. The winter cover crop was seeded during the first two weeks of September each year and consisted of a winter triticale/hairy vetch mix. The crop was harvested mid-June each year for hay. Following hay harvest, the fields were burned down with Glyphosate and seeded to a 7-way cover crop mix (**Table 2**) that was harvested with yearling beef cattle steers. Plant root diversity as well as plants that form association with AMF are important for inclusion in cover crop seed blends. Sunflower, oat, pea, and hairy vetch are crops with roots that develop associations with AMF. Whereas rape, cabbage, and turnip are included in the 7-way mix to provide diversity, nitrogen scavenging, and aid in reducing compaction; however, do not form AMF associations. Fifty percent of the crop species included in the cover crop blend were legumes. The importance of legumes in cover crop mixes cannot be over emphasized, because their nitrogen fixing characteristics provide a nitrogen source for the subsequent crop through microbial nutrient cycling.

The cropping system consisted of crops with small seed size (spring wheat, cover crop, pea, barley) that were seeded using a John Deere 1590 No-Till drill (row spacing: 19.1 cm) and crops with large seed size (corn and sunflower) that were planted using a John Deere 7000 No-Till planter (row spacing: 0.762 m) (Deere & Company, Moline, IL USA). Spring wheat was planted to achieve 3.09 million plants per ha and the corn and sunflower crops were planted to achieve plant populations of 7,692 plants per ha. The mixed crop of field pea (Arvika, var.) was seeded at 67.2 kg/ha and the forage barley (Stockford, var.;) was seeded at 44.8 kg/ha, i.e., 60.0% pea – 40% barley.

Crop	**kg/ha**	**Percent**
Sunflower	10.9	4.01
Oat (var. Everleaf)	108.7	40.01
Winter Pea (var. Flex)	108.7	40.01
Hairy Vetch	27.2	10.01
Forage Rape (var. Winfred)	5.4	1.99
Ethiopian Cabbage	5.4	1.99
Hunter Leaf Turnip	5.4	1.99

Table 2.
Seven-Way cover crop mix.

The order in which crops occurred in the rotation was based on plant season of growth (cool- or warm-season), water use, and residue C:N ratio. Starting with spring wheat as the first crop in the rotation, the crop is rated as medium for water use and was followed by the multi-specie cover crop that contained a high level of legume plants with low C:N ratios. Under normal precipitation in the region the cover crop would be expected to breakdown due to microbial nutrient cycling making plant nutrients available to the subsequent corn crop. Corn is a warm-season grass crop and sunflower is a warm-season broadleaf crop, and both crops are rated as high water use crops; therefore, a cool-season mixed grass and broadleaf intercrop mix (field pea-forage barley) suitable for grazing was placed between corn and sunflower, because both crops in the mix are rated as being low water use crops and the mix was 60% legume. The high concentration of legume in the crop mix with a low C:N ratio was expected to provide nitrogen and other nutrients for the following sunflower crop.

4. Forage grazing sequence and steers

4.1 Forage grazing sequence

Spring seeded annual forages require adequate growing time before grazing suitability is reached; therefore, yearling steers grazed native range pasture until annual forages were ready for grazing. On average, the steers grazed native range for approximately 108 days between the first week of May and mid-August. Western North Dakota native range pasture grass specie composition consists of both cool- and warm-season grasses: cool-season: western wheatgrass (*Pascopynum smithi*), slender wheatgrass (*Elymus trachycaulus*), prairie junegrass (*Koeleria macrantha*), bluebunch wheatgrass (*Pseudorognerta spiacata*), green needlegrass (*Nassella viridula*), slender wheatgrass (*Elymus trachycaulus*), warm-season: prairie sandreed (*Calamovilfa longifolia*), indiangrass (*sorghastrum nutans*), blue grama (*Bouteloua gracilis*), sideoats grama (*Bouteloua curtipendula*), and little bluestem (*Schizachyrium scoparium*). For the research, control groups grazed native range for the full grazing season; however, grazing groups that were assigned to graze annual forages began grazing field pea-barley (*Pisum sativum*, var. Arvika; *Hordeum vulgare*, var. Stockford) as the first annual forage in the grazing sequence followed by corn and cover crop grazing. Crop grazing readiness for pea-barley was determined when the barley grain was in the early milk stage and peas were small and soft (2–3 mm).

4.2 Steers, stocking density, and grazing management

Over the course of multiple experiments steers of differing frame score have been used as grazing animals and described as small frame (SF: Frame Score Range: 3.77–3.82) and large frame (LF; Frame Score Range: 5.53–5.63). Frame score determination is computed according to the formula: $-11.548+(0.4878 * Ht) - (0.0289 * Age) + (0.00001947 * Age^2) + (0.0000334 * Ht * Age)$, where age = days, and height = inches [16]. For one research evaluation [17] only one steer frame score type was used (LF), and in other studies [18, 19] steers of both SF and LF types were used. Grazing equivalents for steers used in these research investigations were computed from a reference cow (454 kg) nursing a six-month old calf [20]. Grazing equivalents for each steer type were calculated by conversion of reference animal cow weight and SF and LF steer weights to metabolic weight, which resulted in grazing steer equivalents of 0.840 and 0.934 for steers categorized as being of SF and LF. Each of the pastures in the grazing sequence were 1.74 ha in size, replicated three times and each pen replicate stocking rate was 0.2138 ha per steer. The field pea-forage barley intercrop mix was grazed for 27–32 days and varied by year. The mixed intercrop maturity progresses rapidly from seeding to full maturity and it was determined that extending the grazing period beyond 27–28 days resulted in a decline in steer gain performance. For some of the studies, the intercrop mix was windrowed to capture forage quality before the onset of grazing and in other studies the crop was grazed as standing crop. Windrowing was not always successful. Above average precipitation one year resulted in moldy feed in the windrows, which was undesirable. Therefore, windrowing was suspended for subsequent research projects. Corn grown for sequence grazing was unharvested (not combined residue) vegetative actively growing plant of a forage-type categorized as being later maturing and used for silage due to the plant's stalk to leaf ratio and soluble sugar content. Days of forage corn grazing ranged from 52 to as much as 71 days and was largely dependent on the amount of rain received. Cover crop mix was the last crop grazed in the sequence and the amount of above ground biomass available for grazing was more variable than the preceding pea-barley and corn crops. The observed variability is directly related to available soil moisture and precipitation following harvest of the winter triticale-hairy vetch cover crop mix. Insufficient soil moisture delayed germination for as much as four to five weeks before precipitation was received, which negatively impacted total above ground biomass for grazing.

Upon completion of sequence crop grazing, bale grazing was initiated. For bale grazing, cover crop hay is produced using a full-season cover crop consisting of oats, peas, sorghum-sudan, and clover (crimson var. and berseem var.). Nutrient analysis of the cover crop bales and the starting and ending forage analysis for the other sequence crops that were grazed are shown in **Table 3**. Native range, field pea-barley, corn, cover crop mix, and cover crop baled hay were analyzed for crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), invitro organic matter

	CP, %	NDF, %	ADF, %	IVOMD, %	IVDMD, %	Ca/Phos, %	TDN, %
Native Range							
Start	9.7	64.7	35.4	57.5	58.7	0.27/0.13	55.5
End	6.9	58.8	38.9	47.4	48.6	0.31/0.11	52.6
Field Pea-Barley							
Start	11.0	55.0	30.2	69.6	68.5	0.50/0.23	59.7
End	8.2	67.0	37.9	54.8	54.1	0.37/0.25	53.5
Corn (Whole Plant)							
Start	7.7	56.6	29.5	78.0	77.6	0.32/0.24	60.1
End	4.6	69.2	38.2	64.7	63.6	0.17/0.20	53.2
Cover Crop							
Start	11.8	50.5	31.5	43.0	69.3	0.75/0.34	58.7
End	12.3	52.8	34.5	64.3	61.9	0.83/0.31	56.4
Cover Crop Bale	12.8	54.4	31.4	72.5	72.3	0.48/0.22	59.0

Table 3.
Forage nutrient analysis for grazing sequence crops and cover crop bales.

disappearance (IVOMD), invitro dry matter disappearance (IVDMD), calcium and phosphorus (Ca/Phos), and total digestible nutrients (TDN).

5. Alternative integrated systems research

Integrated systems research has focused on three primary areas of interest: crop production, beef production, and soil health within the systems' evaluation. The ensuing discussion will look into each area of interest and the complementary aspects of the holistic regenerative approach to the systems' integration.

5.1 Crop production and soil health

The cropping system [21] consisted of hard red spring wheat grown continuously on the same replicated fields for the entire investigation and is designated as HRSW-C. The spring wheat control is compared to hard red spring wheat grown in the five-crop rotation and has been designated as HRSW-R. The continuous spring wheat control is a very important part of the research, because wheat farmers in the region have grown spring wheat on the same land for decades (30 to 50 years). Under these conditions, the only possible way to raise a good crop of spring wheat is to apply fertilizer based on soil test results for a given yield goal (44.8–56 kg/ha). The alternative is to employ a holistic approach that considers the principles of soil health that includes multi-crop diversity and integration of beef cattle grazing. The crop rotation of spring wheat, cover crop, corn, field pea-barley mix and sunflower with three of the rotation crops being harvested by grazing has the potential to reduce the cost of production and enhance profitability. At the start of the research, urea nitrogen fertilizer was applied according to soil test results to both the control and rotation spring wheat fields. The HRSW-R fertilizer application was discontinued after two years and after three years fertilization of the HRSW-C was discontinued. Soil fertility was evaluated by creating root restriction zones in the replicated spring wheat fields using aluminum irrigation pipes (20.3 × 61.0 cm) pressed into the soil with an industrial type construction front-end loader. Soil samples were collected from inside and outside the irrigation pipe restriction zones. Economic analysis was carried out with the assistance of the ND Farm and Ranch Business Management Education program budgets in which actual incurred expenses that include fertilizer, chemicals, seed, and crop insurance premiums were entered into the budgets. From the budgets, calculations for individual crop expense, gross return, and net return were determined. Improving soil health through integration of complementing crop types and cattle grazing reduced reliance on mechanical harvesting while aiding in the enhancement of soil nutrient cycling consuming less fuel and fertilizer, and adding value to yearling steers prior to feedlot entry.

Control and rotation HRSW yield did not differ for the five-year cropping system period between 2011 and 2015. Although there was no difference in grain yield, protein percent or test weight, mean grain yield does not fully explain soil health changes that occurred due to the effect soil microbial nutrient cycling had on nutrient supply without the addition of exogenous N fertilizer.

Corresponding to soil nutrient cycling after N fertilizer (urea) was discontinued resulted in a yield transformation whereby spring wheat-control yield was greater initially (**Figure 1**) followed by a continual decline whereas the soil derived nutrient supply supported continued yield increases (**Figure 2**) years 4 (8.4%) and 5 (32.6%) as depicted by chart trendlines.

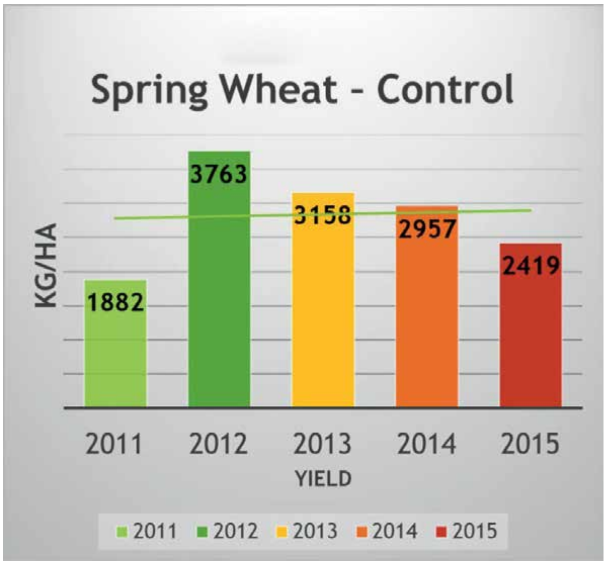

Figure 1.
Spring wheat – control.

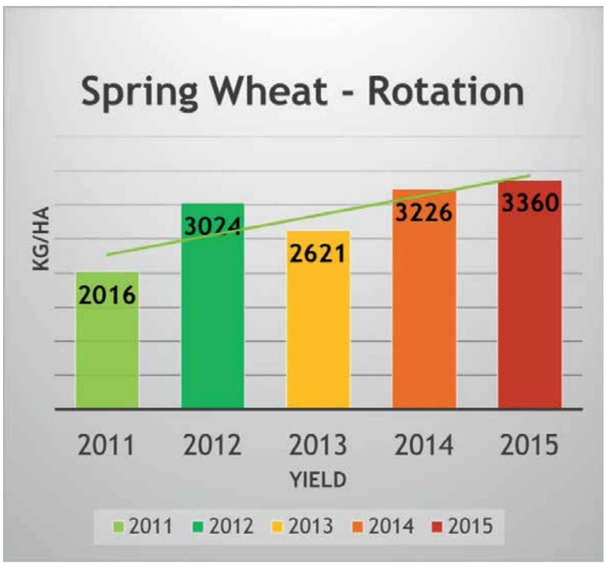

Figure 2.
Spring wheat – Rotation.

Economics for the crop production (**Table 4**) suggest an advantage for the holistic production such that rotation spring wheat had a $6.00/ha greater net return at the time the analysis was performed. The combined net return economic advantage for crops grown in the integrated system was $2,036 compared to $1,514 for the control indicating that although growing spring wheat continuously on the same land year after year requires less intensive management profitability is reduced 34.5%.

Plant diversity within the multi-crop rotation that included spring wheat as well as the other rotation crops (cover crop, corn, pea-barley, sunflower) contributed to

Yield	HRSW-C[1] (kg/ha)	HRSW-R[1] (kg/ha)	WT-HV & CC[1] (T/ha)	CORN SILAGE[2] (T/ha)	PEA BARLEY (T/ha)	SUN-FLOWER (T/ha)
5-yr Avg, (P = 0.30)	2,829	2,856	3.7/6.2	8.6	8.4	1.6
5-yr Average Economic Analysis						
Net Return/ ha, $	78.8	95.2	69.22	86.58	110.32	169.12
System Net Return, $[3]	$1,696			$2,279		

[1]*HRSW-CON: Hard Red Spring Wheat – Control; HRSW- ROT: Hard Red Spring Wheat – Rotation; WT-HV&CC; Winter Triticale – Hairy Vetch & 7-Specie Cover Crop*
[2]*Corn silage grain content 2011-2015: 941, 3468, 5519, 2822, 4930 kg/ha (Avg. 3536 kg/ha)*
[3]*Average total 5-year net return for HRSW-C and rotation crops (HRSW- ROT, WT-HV&CC, Corn Silage, Pea Barley, and Sunflower)*

Table 4.
Five-year crop yields and system net return (2011–2015).

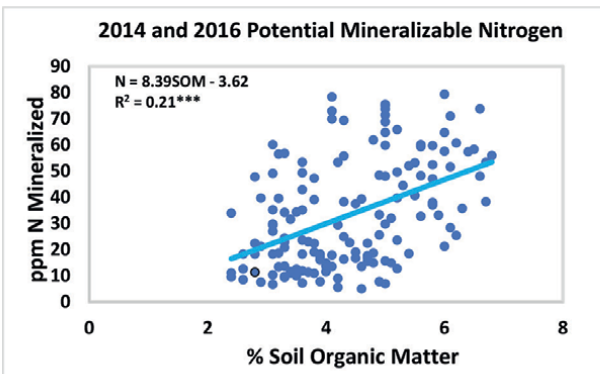

Figure 3.
2014 and 2016 potential mineralizable nitrogen.

an increase in soil organic matter (**Figure 3**.) in the experimental fields that ranged from 2.8 to 6.8% by the end of the five-year period. Contrasting percent SOM with potential N mineralization using regression analysis identified that as the percent of SOM increased there is approximately 8.4 mg N mineralized for each 1.0% SOM increase per kg of soil [22].

Paralleling the systems evaluation were soil properties of interest (water infiltration, wind erodibility, and water stable aggregates). Following five years of cropping history the crop rotation system has numerically greater water stable soil aggregates, reduced potential wind erodibility, and water infiltration rate levels increased (27.1 mm/hr. vs. 19.1 mm/hr.) in the crop rotation indicating that the multi-crop system has a positive effect on soil health.

5.2 Beef production and delayed feedlot entry

The stressors of commingling, transportation, change of feed, new location and dehydration coupled with less developed immune defenses make young cattle less resilient to respiratory pathogen challenges from BRD than older animals with more developed immune systems when challenged by viral and microbial pathogen invasion. Feedlot BRD disease occurrences have been categorized into cohort groupings as being either early-, mid-, or late-feeding stage occurrences that coincide with the first 42-days on feed (DOF), 43–71 DOF, and 72–100 DOF in a mid-western feedlot where mid- and late-stage risk for BRD incidence was evaluated. Incidence for BRD was determined to be greater during the second quarter of the year which coincides with wide temperature fluctuations, summer heat, and humidity [23]. This data set can be contrasted with economist's evaluation [24] using pen-level data (5,773 pens, 636,042 head received) from a Southern Great Plains feedlot where a 2.28% death loss was identified. Sixty percent of the cattle were sourced from auction sale barns. Risk factors for sickness and death loss include sourced from sale barns, travel distance and animal shrink greater than 5.5%, and larger pen size. Customer owned cattle sourced directly from ranches had lower death loss of 1.97% compared to 2.35% among feedlot company owned sale barn sourced cattle. The data also identified that pens of cattle with lighter feedlot arrival in-weights have higher death loss such that for each in-weight increase of 45.4 kg death loss was reduced by 0.2%. Using this percent death loss age reduction statistic, delaying feedlot entry until steers and heifers enter the feedlot weighing 454 to 499 kg (1,000 to 1,100 lbs.) could potentially reduce death loss by 1.1 to 1.3%. Yearling steers involved in integrated crop-beef cattle extended grazing delayed feedlot entry systems research discussed herein are seven to eight months older and 188 to 210 kg heavier upon feedlot arrival than cattle in the Southern Great Plains feedlot data set. Due to greater feedlot arrival weight in the delayed feedlot entry research, the steers reach harvest target condition after 82 DOF, but are not immune from BRD and digestive health death loss. However, death loss is substantially reduced. During the 8-year period (2013–2021), death loss for delayed feedlot entry steers was: BRD 0.86%, bloat 0.35%, and unknown 0.17% for a combined 8-year total of 1.38%. In addition, non-performing "realizer" steers were sold at auction for a net revenue loss.

In addition to managing animal health and death loss by withholding steers and heifers from feedlot confinement in retained ownership extended grazing growing systems, the extended grazing program must be profitable. Integrating yearling steers into perennial and annual forage grazing protocols have been studied among differing steer groups with different research objectives [17, 18]. For the initial investigation [17] a control group of randomly assigned steer pen replicates were delivered to the feedlot (FLT: 367 kg In-Weight) and were compared to randomly assigned steers that grazed perennial pasture only (PST) and a third group that grazed perennial pasture and annual forages grown in the diverse multi-crop rotation (ANN). The initial integrated systems investigation objectives were designed to determine 1) the number of days grazed and steer performance, 2) the effect of grazing system on live animal muscle area, fat depth, and intramuscular fat change, and 3) the effect of system on delayed feedlot entry growth performance, carcass measurements, and long-term risk analysis. All steers were grown during the fall-winter-early spring period for modest gain $\leq$ 0.454 kg per steer per day. Grazing start weights for the PST and ANN steers was 369 and 375 kg and ending weights were 509 and 558 kg, respectively. PST and ANN steers gained 140 and 183 kg costing $1.12 and $1.30 per kg of gain. The cost per steer was greater for the ANN steers due to farming costs ($157.31 vs $238.46). Grazing live animal muscle and fat measurements for *longissimus dorsi* muscle area (Ribeye Area; cm^2), backfat depth (cm), and intramuscular fat percentage were monitored as the steers grew grazing perennial and annual forages. Ribeye area for ANN system steers was greater (P = 0.04), fat depth did not differ (P = 0.33), and percent intramuscular fat was 0.70% greater (P = 0.001) (Aloka SSD-500V Portable Ultrasound Machine affixed with Aloka UST-5044–3.5 Linear Array Transducer and Standoff, Sentinel Imaging Group Inc.). The PST and ANN grazing groups grazed for a period of 181 days before transfer to the feedlot for finishing. Feedlot days on feed were longest for the FLT control group (142 days), 91 days for the PST group, and 66 days for the ANN integrated system steers. Compared to the FLT control steers starting and ending weight for the PST and ANN steers was naturally greater due to grazing weight gain. Comparing the three treatment groups in the feedlot, there were no differences measured for ADG, dry matter feed intake (DMI), gain to feed ratio (G:F), and feed cost per kg of gain. Control FLT steer cost was $578.30 compared to $276.12 and $381.18 for the ANN and PST, respectively. Carcass measurements were unremarkable for hot carcass weight (HCW), fat depth (FD), marbling score (MS), USDA yield grade (YG), and quality grade (QG). Upon conclusion of this study the cattle market experienced a down turn in commodity price resulting in undesirable net return values for the FLT control that lost −$298 per steer, PST group that lost −$30.10 per steer, and the ANN grazing system steers that netted $9.09 per steer; a margin of $307.09 between the FLT control and the ANN grazing system steers. A ten-year feedlot sensitivity analysis for the period between 2003 and 2012, and hedging against catastrophic loss was conducted. The sensitivity valuation determined that within the ten-year period the FLT control treatment underperformed seven out of the ten years evaluated. Considering the three treatments FLT, PST and ANN, hedging loss protection was rewarding forty, thirty, and twenty percent of the time. This initial investigation evaluating delayed feedlot entry provided positive direction for future investigations into the potential for managing annual forage crop-grazing systems simultaneously.

Sustaining profitability in the cattle business is not easy. Cow-calf producers generate new wealth when calves are born and subsequently marketed, and the entire beef cattle industry in one way or another receives its livelihood from calves born and reared on ranches across the United States. The rancher, therefore, has direct control over mitigating risk by creating greater beef value before the first point of sale. Resource management and retaining ownership coupled with a vertically integrated business model are powerful tools for creating added beef value. Extracting as much beef value from the cow herd that is practically possible begins with matching cow size and yearling steers of differing skeletal frame-size to the range and annual forage resource.

For the second research project in the series of integrated systems investigations [18], the relationship between cow and steer frame-size, performance, market timing, and economics was evaluated. Rearing environment has a profound effect on cow efficiencies. Brood cow biological efficiency is a complex balance of environmental impact resulting from available feed resources, and interaction between cow frame-size, reproductive efficiency, milking ability, and growth performance [25, 26]. The underlying research premise was that a marketing bias towards calves from small-framed cows exists and profitability at the first point of sale is diminished. Our research team hypothesized that in lieu of selling small-framed calves at weaning using a vertically integrated business model, extended grazing of annual forages, and delayed feedlot entry would eliminate market bias and increase profitability. Yearling crossbred steers (n = 288) from small-framed cows (Aberdeen Angus (Lowline) × Red Angus × Angus × Angus) and moderate to large framed cows (Red Angus × Angus × Simmental × Gelbvieh) were randomly assigned to frame-size groups identified as small-frame (SF) and large-frame (LF) treatment groups. One-half of the frame-size groups were identified as feedlot control groups (FLT) and the remaining one-half were identified as extended grazing groups (GRZ). The mean frame sizes for the FLT control groups were SF: 3.82 and LF: 5.63, and for the GRZ groups, mean frame sizes were SF: 3.77 and LF: 5.53. The FLT control steers were on feed for 218 days compared to 212 days of grazing and 82 DOF in the feedlot for the GRZ treatment steers. When assessing SF steers under grazing conditions compared to their larger framed counterparts, growth was less pronounced; however, the cost per kg of gain was 7.8% less. Beef cattle genetics are constantly improving growth performance and efficiency, and are based on gain test evaluations in which high energy grain-based diets are fed. Therefore, grazing steers consuming forage-based diets are unable to express their full genetic potential for growth. Nonetheless, steers grazing perennial and annual forages grow structurally prior to feedlot entry followed by a compensatory growth and efficiency response in the feedlot when high energy grain-based diets are fed. The SF and LF grazing steers grew at the fastest rate of gain in the feedlot (SF: 1.74, LF 2.10 kg/day) compared to feedlot control SF and LF steers (SF: 1.33, LF: 1.56 kg/day) ($P = <0.01$) and there was no difference in gain to feed efficiency ($P = 0.59$). Total feedlot cost per kg of gain was markedly lower for the grazing steers (SF: $1.53, LF: $1.44/kg of gain) compared to the feedlot control steers (SF: $1.97, LF $1.99/kg of gain). Hot carcass weights for the LF graze and FLT control were 423 and 398 kg, respectively, and hot carcass weights for the SF graze and FLT control were 374, and 350 kg, respectively. Systems economic analysis using a vertically integrated business model from birth to slaughter is shown in **Table 5** that summarizes annual cow cost and steer expenses returns for winter growing and extended grazing, feedlot expenses, and carcass value for the comparative frame score groups in the FLT and GRZ systems' treatments.

Item	FLT[2] (LF)[3]	FLT[2] (SF)[3]	GRZ[2] (LF)[3]	GRZ[2] (SF)[3]	SEM	P-Value Trt
Cow, calf wintering, & grazing cost, $	755.51	630.64	1040.67	868.75		
End steer market value, $			1570.45[a]	1553.35[b]	7.37	0.01
Net return/steer, $			529.78	684.60		
Net return/ha, $[4]			26.03	36.71		
System Expenses:						
Cow, calf wintering, grazing, & feedlot finishing /steer, $	1452.74	1222.74	1312.09	1107.56		
Income						
Carcass value/ steer, $	2042.47	1753.88	2243.61	2017.51	91.81	0.79
System net return/steer, $	589.73	531.14	931.52	909.95		

[a–b]*Means with different superscripts within a line are significantly different, ($P \leq 0.05$).*
[1]*3-Year mean*
[2]*FLT: control steers moved directly to the feedlot for growing and finishing; and GRZ: steers grazed a sequence of native range, field pea-barley, and unharvested corn before transfer to the feedlot at the University of Wyoming*
[3]*SF: Small Frame, LF: Large Frame*
[4]*Net return/ha based on sum of native range and annual forage hectares grazed per steer*

Table 5.
Effect of grazing and retained ownership vertical integration on net return[1].

At the end of the 212-day grazing period, the yearling steers were valued, but not sold to establish an end grazing steer value and calculate net return per ha values before transfer to the finishing feedlot. Small-frame steers cost less to produce and had greater grazing net return per ha. Due to lower placement cost and total system expense, the SF grazing steers cost less to produce and compared to the LF grazing steers that had the highest net return the SF grazing steer net return was a mere 2.32% less. Upon further inspection, comparing the SF grazing steer net return to the SF feedlot steer net return, the SF grazing steer net return was 41.63% greater illustrating the effect that extended pre-feedlot grazing and compensating feedlot gain can have on system net return.

Frame-size evaluation shown here clearly identifies that beef cattle producers in semi-arid regions can maintain cows with smaller frame-size taking advantage of increased stocking rate and greater net return per ha per cow exposed and eliminate calf market bias through retained ownership in a vertically integrated business model from birth to final harvest.

For the third study in the series of investigations into to evaluating extended grazing and delayed feedlot entry [18], the question was asked, "Will withholding yearling steers from feedlot confinement through grazing above average quality cover crop hay after integrated systems grazing has been completed be more profitable than grazing native range only?" Feeding large round hay bales weighing 499 to 635 kg (1,100 to 1,400 lbs.) in spacious non-confined areas was previously described as "bale grazing". Using the same integrated systems research infrastructure protocol and economic analysis previously defined, replicated groups (3 reps) of yearling steers grazing native range only were compared to replicated groups grazing a sequence of native range and annual forages (pea-barley, corn, and cover crop) was the foundation for the 3-year project. As such, when NR and the sequence of NR and ANN forage grazing was completed bale grazing started. The seasonlong cover crop fed was seeded in May each year consisting of Pea, barley, sorghum-sudan hybrid, crimson clover, and berseem clover and harvested to obtain hay with crude protein value ranging from 12–14% CP. **Table 3**, shows the nutrient analysis of the cover crop hay that had a crude protein value of 12.8% and Total Digestible Nutrient value of 59.0%. Bale grazing withheld the steers from feedlot confinement for an additional 43.7 days. The combination of sequence forage grazing and the additional time steers spent grazing bales resulted in a 43.0 kg weight advantage compared to the NR control steers, which carried through to the end of the finishing period. Gross carcass value over the three-year period of the study was $92 greater (P = 0.031) than the NR steers ($1,922 vs $2,014). During the three-year study and economic analysis, ANN forage sequence steers were consistently heavier entering the feedlot and the grazing weight margin gained between the NR control steers and the ANN forage sequence steers did not change appreciably during feedlot finishing resulting in ANN forage system steers fed harvested baled hay before feedlot entry being consistently more profitable.

6. Conclusion

Confining cattle in close proximity to each other greatly increases social stress and animal-to-animal disease transmission. Reducing the use of antibiotics in growing and finishing beef cattle is impossible without significant modification in beef cattle management before confinement feedyard placement. Non-confinement investigations incorporating crop production and beef cattle grazing reported herein have defined successful protocols that reduce the need for using antimicrobials for animal well-being from minimal at weaning to nearly non-existent during grazing. Non-confinement protocols have increased utilization of home-grown crops through crop and beef cattle grazing integration and by employing the five principles of soil health. Employing a diverse multi-crop rotation with beef cattle grazing increased water infiltration and soil water holding capacity as well as improved and reduced reliance on commercial fertilizer application. Additional merits of the systems' integration are found in improved wildlife habitat for birds, small animals, and large game. Beef cattle grazing a sequence of perennial and annual forages improved grazing animal performance, carcass weight and net return. Reducing agronomic inputs that can be replaced with naturally occurring nutrient cycling has the potential to increased profitability for the farm-ranch enterprise and improve revenue for local economies. Moreover, systems analysis has shown that significant modifications to cattle management before the first point of sale using retained ownership and an annual forage grazing integration protocol from birth to slaughter can be profitable for the cow-calf producer. Well in advance of any business model transition to an integrated crop-livestock system from birth to slaughter, ranch managers considering a business model shift are encouraged to conduct an in-depth through feasibility analysis of the proposed change and cash flow that will support the enterprise modification to include infrastructure additions for cropland fencing and water installation as well as establishing bank operating loan repayment schedule during the business model transition.

Conflict of interest

The authors declare no conflict of interest.

Author details

Songul Senturklu[1], Douglas Landblom[2*], Gerald Stokka[3] and Larry Cihacek[4]

1 Canakkale Onsekiz Mart Universities, Canakkale, Turkey

2 North Dakota State University – Dickinson Research Extension Center, Dickinson, North Dakota, USA

3 Department of Animal Science, Extension Veterinarian/Livestock Stewardship Specialist, North Dakota State University, Fargo, North Dakota, USA

4 School of Natural Resource Sciences, Soil Scientist, North Dakota State University, Fargo, North Dakota, USA

*Address all correspondence to: douglas.landblom@ndsu.edu

References

[1] Duff GC, Galyean ML. Board-invited review: Recent advances in management of highly stressed, newly received feedlot cattle. Journal of Animal Science. 2007;**85**:823-840

[2] Nickell JS, White BJ. Metaphylactic antimicrobial therapy for bovine respiratory disease in stocker and feedlot cattle. The Veterinary Clinics of North America. Food Animal Practice. 2010;**26**:285-301

[3] Taylor JD, Fulton RW, Lehenbauer TW, Step DL, Confer AW. The epidemiology of bovine respiratory disease: What is the evidence for preventive measures? The Canadian Veterinary Journal. 2010;**51**:1351-1359

[4] Mcartney JE, Bateman KG, Ribble CS. Health performance of feeder calves sold at conventional auctions versus special auctions of vaccinated or conditioned calves in Ontario. Journal of the American Veterinary Medical Association. 2003;**223**:677-683

[5] Lewis JM, Klopfenstein TJ, Stock RA. Effects of rate of gain during winter on subsequent grazing and finishing performance. Journal of Animal Science. 1990;**68**:2525-2529

[6] Shain DH, Klopfenstein TJ, Stock RA, Viselmeyer BA, Erickson GE. Evaluation of grazing alternative summer and fall forages in extensive beef cattle production systems. Journal of Animal Science. 2005;**21**:390-402

[7] Sindt M, Stock R, Klopfenstein T. Calf versus yearling finishing. In: Nebraska Beef Cattle Report. Lincoln: University of Nebraska; 1991. pp. 42-43

[8] National Research Council. Predicting Feed Intake of Food Producing Animals. Washington, DC: National Academy Press; 1987. pp. 61-62

[9] Cabrera J, Estrada R, Delagarde P, Faverdin J, Peyraud L. Dry matter intake and eating rate of grass by dairy cows is restricted by internal, but not external water. Animal Feed Science and Technology. 2004;**114**:59-74

[10] National Research Council. Nutrient Requirements of Beef Cattle: Factors Affecting Feed Intake. 7th ed. Washington, DC: National Academy Press; 1996. pp. 85-86

[11] Senturklu S, Landblom DG, Paisley S, Stroh C. Effect of feedlot finishing compared with grass fed beef following grazing and delayed feedlot entry on steer performance, carcass measurement and net return. Journal of Animal Science. 2022;**2022**:186

[12] U.S. Climate Data. Available from: https://www.usclimatedata.com/climate/dickinson/north-dakota/united-states/usnd0090 [Accessed: July 29, 2022]

[13] United States Department of Agriculture/Burleigh County Soil Conservation District, Menoken Farm. Five Soil Health Principles. https://www.menokenfarm.com [Accessed: January 29, 2022]

[14] United State Department of Agriculture – Natural Resource Conservation Service. Practice code 340. Available from: https://www.nrcs.usda.gov/wps/portal/nrcs/detail/national/programs/financial/eqip/?cid=nrcs144p2_027123 [Accessed: August 01, 2022]

[15] Magdoff F, Van Es H. Building Soils for Better Crops: Ecological Management

for Healthy Soils. 4th ed. College Park: Sustainable Agriculture Research & Education; 2021. pp. 138-139

[16] Guidelines for Uniform Beef Improvement Programs. Beef Improvement Federation, 9th ed. 2016. Available from: chrome-extension://efaidnbmnnnibpcajpcglclefindmkaj/https://beefimprovement.org/wp-content/uploads/2013/07/BIFGuidelinesFinal_updated0916.pdf [Accessed: August 01, 2022]

[17] Senturklu S, Landblom DG, Maddock R, Petry T, Wachenheim CJ, Paisley SI. Effect of yearling steer sequence grazing of perennial and annual forages in an integrated crop and livestock system on grazing performance, delayed feedlot entry, finishing performance, carcass measurements, and systems economics. Journal of Animal Science. 2018;**96**:2204-2218

[18] Senturklu S, Landblom D, Paisley S, Wachenheim C, Maddock R. Frame score, grazing and delayed feedlot entry effect on performance and economics of beef steers from small- and large-framed cows in an integrated crop-livestock system. Animals. 2021;**11**:3270

[19] Senturklu S, Landblom DG, Paisley S. Economic impact of bale grazing on delayed feedlot entry steer performance, carcass measurement, and net return. Journal of Animals Science. 2021;**99**:97

[20] Sedivec KK, Printz JL. Ranchers Guide to Grassland Management IV. North Dakota, USA: North Dakota State University; 2014. pp. 44-45

[21] Landblom DL, Senturklu S, Cihacek L, Brevik E. Effect of a multi-crop rotation on soil health, spring wheat and rotation crop yields, and cropping system economics in western North Dakota. In: Dickinson Research Extension Center Annual Report. Dickinson, North Dakota, USA: NDSU Dickinson Research Extension Center; 2016. Available from: https://www.ag.ndsu.edu/dickinsonrec/archive/annual-reports-1/2016-annual-report/effect-of-hrsw-5crop-rotation-crp-prod-v4-final-8.pdf

[22] Cihacek L, Senturklu S, Landblom D. Mineral N cycling in an integrated crop-grazing system. In: Dickinson Research Extension Center Annual Report. Dickinson, North Dakota, USA: NDSU Dickinson Research Extension Center; 2019. Available from: https://www.ag.ndsu.edu/dickinsonrec/archive/annual-reports-1/2019-annual-report/nitrogen-mineralization-cihacek-et-al.pdf

[23] Smith KJ, Amrine DE, Larson RL, Theurer ME. Determining relevant risk factors associated with mid- and late-feeding-stage bovine respiratory disease morbidity in cohorts of beef feedlot cattle. Applied Animal Science. 2022;**38**:373-379

[24] Buda M, Curry-Raper K, Riley J, Peel D. Determinants of feedlot cattle death loss rates. Journal of Applied Farmer Economy. 2020;**3**:1-10

[25] Cartwright TC. The use of systems analysis in animal science with emphasis on animal breeding. Journal of Animal Science. 1979;**49**:817-825

[26] Jenkins TG, Ferrell CL. Productivity through weaning of nine breeds of cattle under varying feed availabilities: I. Initial evaluation. Journal of Animal Science. 1994;**72**:2787-2797

Chapter 7

Mitigation of Environmental Impact of Intensive Animal Farming through Conversion of Animal Wastes to Value-Added Products

Abigail N. Tasaki and Ken Tasaki

Abstract

The environmental impact of concentrated animal farming operations has become serious social issues, with the livestock wastes contaminating waterways and groundwaters and generating greenhouse gas (GHG) emissions that are responsible for more than half the total GHG emissions in agricultural activities in the U.S. These impacts are mostly due to the current practice of spraying manure or manure digestate on croplands. We have recently developed two novel processes not only to mitigate the impacts stemming from the current manure management practice but also to bring in extra revenues to livestock farmers, which should provide an incentive to the farmers, by recovering value-added products from livestock manure or manure digestate. In this review, we discuss the effectiveness of the processes to produce two products: protein hydrolysate feed additives from the manure-digestate solid by one process and renewable ammonia from the manure-digestate liquid by another. One process uses thermal hydrolysis to extract protein from manure-digestate solid at a moderate recovery rate of more than 60%. Another employs acid-base reactions to strip NH_3 from manure-digestate liquid and dissolve the stripped NH_3 gas into the water at a high recovery rate of 90%. By repeating this stripping process, the nitrogen concentration in the water can reach as high as 18%.

Keywords: dairy digestate, greenhouse gas, renewable ammonia, protein hydrolysate, thermal hydrolysis

1. Introduction

According to Food Agriculture Organization, the world's meat consumption in 2020 was estimated to be 328 Mt and is expected to reach 374 Mt by 2030 [1]. As the meat consumption increases worldwide, the environmental impacts of livestock agricultural wastes are becoming serious issues. Insufficient disposal or management of the wastes can not only result in environmental contaminations, but also create human health hazards. Inadequate manure management has been causing eutrophication in rivers, lakes, and bays through runoff of excess nutrients as well as groundwater contamination by leaching of manure through the soil. As a result, livestock manure alone is responsible for about 100 $TgNy^{-1}$ of the nitrogen (N) input on the earth in 2005, twice as much as the N input of roughly 50 $TgNy^{-1}$ in 1950 [2].

Anaerobic digestion of livestock manure is becoming popular, driven by an incentive to receive the renewable identification number by selling biomethane. An anaerobic digester (AD) generates the digestate as the discharge which is often treated by a solid-liquid separation. The solid is often used as low-valued solid fertilizers, while the liquid is sprayed on croplands. This N-rich liquid needs to be sprayed over a wide area to keep the nitrogen level in the soil at a certain level, currently regulated at the state levels. As the consolidation of livestock operations progresses, the livestock headcount/farm grows rapidly, generating ever more manure per ft^2. As a result, regulations as to how much N can be sprayed per ft^2 of croplands are also being tightened. As a consequence, it is becoming increasingly difficult for livestock farmers to apply manure liquid economically without exceeding the N nutrient required for growing crops in the nearby lands in order to avoid the build-up of excess N in the soil.

The current practice of applying the digestate (to be referred to as digestate) solid and liquid can cause another problem: greenhouse gas (GHG) emissions. Nitrous oxide (N_2O), almost 300 times as potent as the global warming potential of CO_2, is generated by anaerobic denitrifying bacteria in the soil from ammonia after fertilizer applications. More than half (54.8%) of the total GHG emissions from agricultural activities in the U.S. are due to the N_2O emissions from fertilizer applications to the soil. Of those N_2O emissions, 30–50% originate from applications of animal manure, which includes organic nitrogen [3, 4]. Application of liquid manure or digestate to the soil provides the available N and carbon, which in turn promote heterotrophic activity, depleting the oxygen availability in the soil, and thus favor the creation of anaerobic microbes that release N_2O via denitrification [5, 6]. The current manure management practice causes the disruption of the N cycle on Earth. However, there is potentially a considerable opportunity to reduce the N_2O emissions from the soil without spraying manure as is, but instead by applying clean inorganic nitrogen recovered from animal manure without creating anaerobic conditions in the soil. This approach will be the first focus of our report.

Table 1 lists the average concentrations of various nitrogen sources determined by daily samplings by one of the authors over a week from the digestate liquid after a solid-liquid separation on a dairy farm with 5000 heads in a Midwestern state. As **Table 1** indicates, the concentration of NH_4^+ is the highest among nitrogen sources. Since NH_4^+ can be easily converted to NH_3, depending on the temperature and pH, a significant volume of NH_3 can be lost through emissions into the atmosphere during the storage in a lagoon, though some go through the natural transformation of

Nitrogen	Concentration
TN[a]	1900
NH_4^+	1800
NO_3	9
N_{org}[b]	91

[a]*The total nitrogen.*
[b]*The organic nitrogen.*

Table 1.
Nitrogen concentrations in dairy digestate liquid (mg/L).

nitrification or denitrification during the storage as well. Therefore, it is preferable to recover NH_4^+ before storage in a lagoon.

The nutrient runoff or leaching of nutrients in manure into the soil is also a waste of agricultural resources. Livestock manure is an important source of nutrients for crops and grains. For example, the U.S. annual consumption of N for crop production was 13 Mt in 2015 [7]. On the other hand, the estimated N produced from livestock animal manure in 2007 in the U.S. was 6.2 Mt [8]. Almost half the N fertilizer consumption for crop productions could be replaced by N recovered from animal manure. Accordingly, the recycling of N from manure is a key to the efficient utilization of agricultural resources and the protection of the environment.

Stripping/scrubbing processes are common for the NH_3 recovery. There are many NH_3 stripping and scrubbing processes such as AMFER [9], Dorset LGL [10], and BIOCAST Process [11]. AMFER can have a relatively high NH_3 recovery rate of 80%; yet it uses heat for the NH_3 stripping which increases the operation cost. What is common among these ammonia stripping processes is that the initial and operational costs of ammonia recovery using a stripping tower and a scrubber tower are high in general. The CAPEX has been estimated to be up to $17.5 million for 800 m^3 day^{-1} of flow rate with 2500 NH_4-Nmg L^{-1} of the NH_4^+ concentration [9]. Further, to be cost-competitive with other recovery technologies, the NH_4^+ concentration in the influent must be higher than 2000 mg L^{-1} which limits the application of these processes. In addition, when the digestate liquid is used as the feed, it always contains CO_2 which needs to be removed by a CO_2 stripper prior to the NH_3 stripping, since CO_2 is acidic, raising the caustic soda consumption to increase the pH for the NH_3 stripping. Moreover, the NH_4^+ concentration in the recovered solution is determined by the initial concentration of NH_4^+ in the feed which limits how high the N concentration in the recovered solution can go. As we will explain later, our process can produce high N-concentration fertilizers.

Membranes are often used for NH_3 recovery from manure. Membrane filtrations include ultrafiltration, nanofiltration, reverse osmosis (RO), electrodialysis, and membrane distillation [9]. For example, Riaño et al. have developed a gas permeable membrane to recover NH_3 from manure in a lagoon, controlling the pH gradient between inside and outside of the membrane as the mass transfer driving force [12]. Though high total ammonia removal rates, 79–99%, were obtained, the NH_4^+ concentration in the recovered tank was low, below 2% [12]. Furthermore, all membrane technologies have a fundamental problem of membrane fouling, especially treating manure which has a large content of organic matters, a main cause of fouling.

A challenge of recovering ammonia from manure/digestate liquid is removing undesirable materials in manure slurries without serious membrane fouling or high energy costs and then concentrating ammonia without using high-energy processes such as RO. On the other hand, once NH_4^+ in the manure liquid is transferred to the NH_3 gas, the NH_3 recovery becomes less problematic, leaving behind undesirable materials in the liquid phase. We have developed a simple process to recover NH_3 from manure/digestate liquid by applying acid-base reactions for the NH_3 stripping and dissolving the stripped NH_3 gas into the water.

As to the digestate solid, it contains a considerable amount of protein, from 12 to 48 wt.%, depending on the animal and the growth period, according to the report by Pacific Northwest National Laboratories [13]. There is a substantially large volume of protein in livestock manure that could be potentially recovered, as is shown in **Table 2**.

Currently, such a protein is often being wasted. When the digestate solid is sprayed on croplands, protein in manure tends to stay in the soil longer since protein is not available to plants immediately as a nutrient; hence, it can be subject to environmental

	Protein[a] %	Manure[b] kg/day/head	Solid[c] %	Dry manure[d] kg/day/head	Protein[e] kg/y/head	Head counts[f] million	Protein[g] MMt/y
Dairy	18.1	25.8	12.7	3.3	216.7	144	31.2
Cattle	12.1	22.5	11.6	2.6	138.6	987	136.8
Hog	25.1	4.7	9.2	0.4	36.4	654	23.7
Poultry	39.8	0.1	25.2	0.0	2.1	68,566	146.8
Total							338.7

[a]*The protein content in manure on a dry matter basis [13].*
[b]*The weight of manure discharged a day per head of an animal [14].*
[c]*The solid content in manure [14].*
[d]*The weight of dry manure discharged a day per head of an animal [14].*
[e]*The weight of protein discharged a day per head of an animal.*
[f]*The global head counts of each livestock animal [15].*
[g]*The annual weight of protein discharged by livestock animals globally in million.*

Table 2.
The estimated global volume of protein generated by livestock manure.

contamination before the complete breakdown of protein, if not properly treated. From a point of view of biological wastewater treatments, protein belongs to what is called biologically non-degradable organic nitrogen compounds which are difficult to treat by conventional treatment processes [16]. It would be beneficial if the protein is recovered from manure before it causes environmental problems. The recovered protein can be converted to various value-added products. We propose one application using the recovered protein: an antioxidant feed additive. The protein recovery from manure will be the second focus of this review.

A process of protein extraction from manure solid has been patented, using solvent extraction [17]. Their approach applies a high concentration, 1 M, of an alkali to the extraction. Such an approach not only requires separation of the alkali after the extraction, but recycling or disposal of the spent alkali as well. We have developed alternative extraction process, using the thermal hydrolysis process (THP) without the use of any chemical. THP has been applied to the extraction of bioactive compounds from plants successfully [18, 19]. Very few reports have been published on the application of THP for protein recovery from manure/digestate solid in the literature. We will examine the efficacy of the protein recovered from the digestate solid for its antioxidant activity.

Our objective is to discuss the two novel processes to recover value-added products from both digestate solid and liquid not only to mitigate eutrophication and GHG emissions associated with livestock manure, but also to bring in extra revenues from the products, some of which can be a renewable N fertilizer or non-carbon renewable energy, bioammonia, and an antioxidant feed additive.

2. Materials and methods

2.1 Materials

2.1.1 NH_3 recovery

For the NH_3 recovery, we used a formulated dairy digestate liquid sample by preparing an NH_4^+ solution with the NH_4^+ concentration of 2100 mg/L and alkalinity of 8800 mg/L by mixing NH_4OH and $NaHCO_3$, respectively, with 1 L of distilled water. This was to simplify numerous experiments anticipated to be performed to optimize the recovery conditions. Once optimized, the real digestate liquid will be used for validation of our results. The above concentrations were obtained by analyzing samples of the digestate liquid taken from a centrifuge effluent of the dairy digestate on a dairy farm with 5000 Holstein cows. The chemicals used were ammonium hydroxide (28% NH_3 in H_2O, Sigma Aldrich), sodium bicarbonate (($\geqq$99.7%, Sigma Aldrich), sodium carbonate ($\geqq$99.5%, Sigma Aldrich), and sulfuric acid ($\geqq$99.99%, Sigma Aldrich), all without further purification.

2.1.2 Protein recovery

The manure digestate solid (DS) sample was collected from the solid separated by a screw separator from the digestate effluent of AD using as the feed the dairy manure from a dairy farm in California Central Valley. The DS sample had a 50% water content. 24.8 g of DS sample was mixed in 1 L of distilled water and treated by THP without any pretreatment. The chemical composition of the dried DS sample is listed in Section 3.

2.2 Methods

2.2.1 NH_3 recovery

Our process design principle is to keep the process as simple as possible: our NH_3 recovery system consists of two columns: the NH_3 stripping by aeration in one column and the NH_3 dissolution into the water in another.

Figure 1 illustrates the experimental setup for the NH_3 recovery. Namely, our process strips the NH_3 gas from manure/digestate liquid by aeration, using a low-cost chemical, specifically a Brønsted base, in Column a and then dissolves the NH_3 gas into an acidic aqueous solution in Column b to produce a highly concentrated N solution.

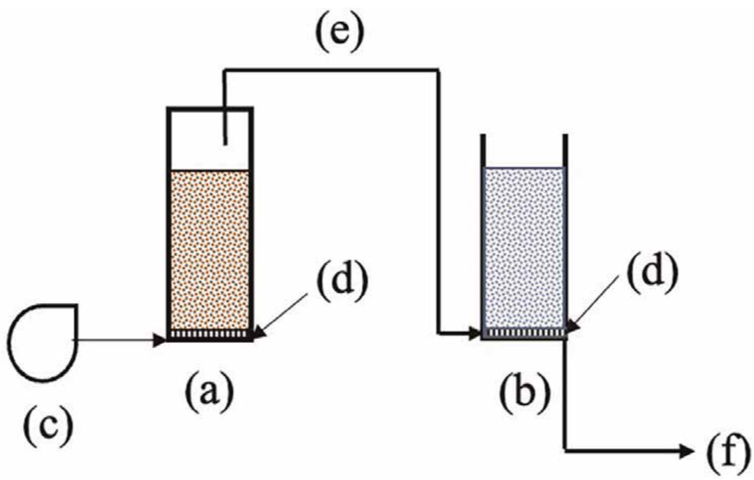

Figure 1.
Experimental setup for the NH_3 recovery: (a) the NH_3 stripping column, (b) the NH_3 recovery column, (c) an air pump, (d) an air diffuser, (e) a pipe, and (f) discharge of the NH_3 solution.

Our process is based on the following reactions for the NH_3 stripping, eq. 1, and the NH_3 dissolving into the water for recovery, eq. 2:

$$NH_4^+ + B^- \rightarrow NH_3 + BH \quad (1)$$

$$NH_3 + AH \rightarrow NH_4^+ + A^- \quad (2)$$

where B^- and AH refer to an anion of a Brønsted base and acid, respectively. There is no membrane and no evaporator involved in our process. Our process can produce highly concentrated N solutions from which liquefied NH_3 can be obtained. Liquefied NH_3 has an energy density twice as much as liquefied H_2 and is receiving increasing attention as the next generation of zero-carbon energy storage or fuel [20].

The formulated sample was first introduced into Column a, the stripping column, while an acid solution was poured into Column b, the recovery column. The acid solution was prepared by mixing 550 g of 99.99% sulfuric acid with 1 L of distilled water. Stone air diffusers were located at the bottom of each column for aeration. Before aeration of the formulated sample, 7 g of Na_2CO_3 was added to Column a which triggered the following reaction:

$$2NH_4^+ + Na_2CO_3 \rightarrow 2Na^{2+} + 2NH_3 + CO_2 + H_2O \quad (3)$$

The stoichiometric amount of Na_2CO_3, given 2100 mg/L of NH_4^+, was 6.18 g. The excess amount of Na_2CO_3 was added to ensure the completion of eq. 3. According to eq. 3, one mole of Na_2CO_3 produces two moles of ammonia, and CO_2 is produced as a by-product. The subsequent aeration stripped ammonia produced by eq. 3. The NH_4^+ concentration was monitored by a UV-vis photo spectrometer (DR 6000 by Hach) over time. The NH_4^+ removal rate, η_{remove}^{NH4}, was defined by the following equation:

$$\eta_{remove}^{NH4} = 100 \times \left\{1 - \left([NH_4^+]_i - [NH_4^+]_f^{strip}\right)/[NH_4^+]_i\right\} \quad (4)$$

where $[NH_4^+]_i$ and $[NH_4^+]_f^{strip}$ refer to the initial and the final NH_4^+ concentration in Column a, respectively.

The NH_3 gas stripped in the stripping column was sent to Column b, along with other gases, N_2, O_2, and CO_2, through a pipe and dissolved into the acid solution through a stone diffuser. When the NH_3 gas contacts the acid solution, the following reaction occurred:

$$2NH_3 + H_2SO_4 \rightarrow (NH_4)_2SO_4 \quad (5)$$

This is an acid-base reaction continuing until all sulfuric acid is consumed. The NH_4^+ concentration in the recovery column was monitored over time. The NH_4^+ recovery rate, $\eta_{recovery}^{NH4}$, was defined by the following equation:

$$\eta_{recovery}^{NH4} = 100 \times \{1 - ([NH_4^+]_i - [NH_4^+]_f^{rec})/[NH_4^+]_i\} \quad (6)$$

where $[NH_4^+]_f^{rec}$ represents the final NH_4^+ concentration in Column b.

The above operation was continued by replacing the spent formulated sample in Column a with a new one and adding the same amount of Na_2CO_3 into the Column a after each batch aeration until the NH_4^+ concentration no longer increased in the Column b. No additional H_2SO_4 was added to Column b. The original amount of

H_2SO_4 added to Column b, 550 g, was determined by the maximum solubility of $(NH_4)_2SO_4$ in water, 744 g/L. When all H_2SO_4 was consumed in Column b, the operation was stopped.

2.2.2 Protein recovery

We have developed a two-heating step process for the extraction of protein from DS by THP. The detailed description of the THP treatment followed by ultrafiltration (UF) for the recovery of protein from DS has been described elsewhere [21]. We have utilized a series of instrumental characterizations of the recovered protein hydrolysates (PHs): sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE), matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectroscopy, and amino acid analysis (AAA). Then, we evaluated the efficacy of PHs as an antioxidant by the in-vitro oxygen radical absorbance capacity (ORAC) measurements. All methods were described in our previous work [21].

3. Results and discussion

3.1 NH_3 recovery

3.1.1 NH_3 removal and recovery rates

Figure 2 shows the NH_4^+ concentrations in the stripping column as a function of time: (a) without and (b) with Na_2CO_3. When Na_2CO_3 was not used, the concentration decreased about by half and then became a plateau. Since the formulated sample had a pH of 8, NH_3 was stripped in the beginning; hence, the NH_4^+ concentration was reduced. As NH_3 was stripped, H^+ was released by NH_4^+, raising the pH of the formulated sample which slowed down the NH_3 stripping which eventually came to

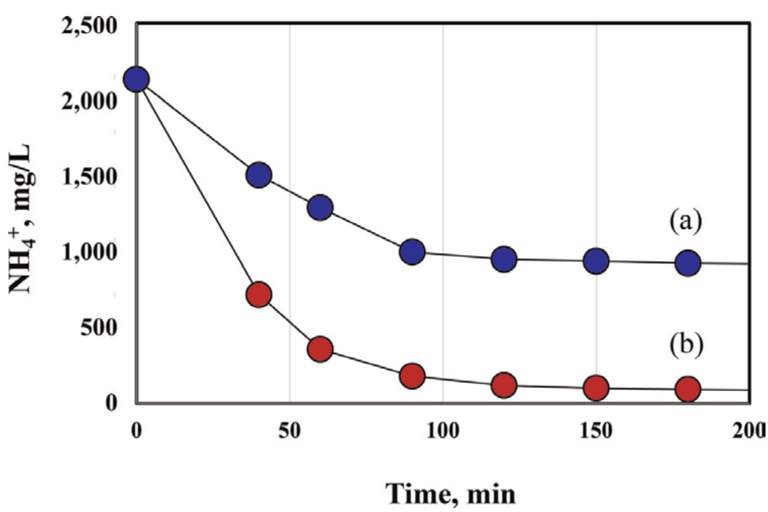

Figure 2.
The NH_4^+ concentrations in the stripping column as a function of time: (a) without Na_2CO_3 and (b) with Na_2CO_3.

an end. When Na_2CO_3 was added, however, the aeration kept stripping NH_3, decreasing the NH_4^+ concentration to nearly zero, driven by eq. 3.

The values of η_{remove}^{NH4} were 96 and 61% with and without Na_2CO_3, respectively. How fast the NH_4^+ concentration decreases depends on the reaction kinetics and the diffusion of the NH_3 gas through the formulated solution to reach air bubbles for the initial NH_4^+ concentration and the volume of the formulated sample.

Figure 3 displays the NH_4^+ concentrations in the recovery column (a) with and (b) without sulfuric acid. A significant difference between the two cases was observed. Without sulfuric acid, the NH_4^+ concentration quickly reached a plateau and did not increase much. CO_2 generated by eq. 3 decreased the pH of water in the recovery column somewhat, dissolving the NH_3 gas into the water to a point; however, the NH_3 dissolution is limited, determined by thermodynamics through pH and the temperature. With sulfuric acid added, the NH_4^+ concentration kept increasing, driven by eq. 5, NH_3 stripped by aeration in the stripping column was mostly recovered in the recovery column. The values of $\eta_{recovery}^{NH4}$ were 90 and 26.9% with and without sulfuric acid, respectively. The efficiency of using chemicals for the NH_3 recovery is clear.

While it took about 2 hours to remove most of NH_4^+ in the stripping column, it took almost 8 hours to dissolve the same amount of NH_4^+ in the recovery column when the volume of water was 1 L for both columns. The reason for the slow process of the NH_3 recovery, relative to the rate of NH_3 removal, is due to the limited amount of the NH_3 gas going into the recovery column available for eq. 3. That is, the flow rate of the NH_3 gas going into the recovery column is smaller than the rate of the NH_3 gas generated in the stripping column through eq. 3. Eq. 3 occurs almost instantly, while eq. 5 only undergoes as the stripped NH_3 gas has a contact with the acidic solution in the recovery column. The rate of eq. 5 is determined by the airflow rate of the air pump, the air flux associated with the air diffusers used, the size of air bubbles, and others. Namely, the faster the rate of the NH_3 gas flow into the recovery column becomes, the more rapidly the dissolving process gets. For example, employing multiple pipes going from the stripping column to the recovery column can speed up the

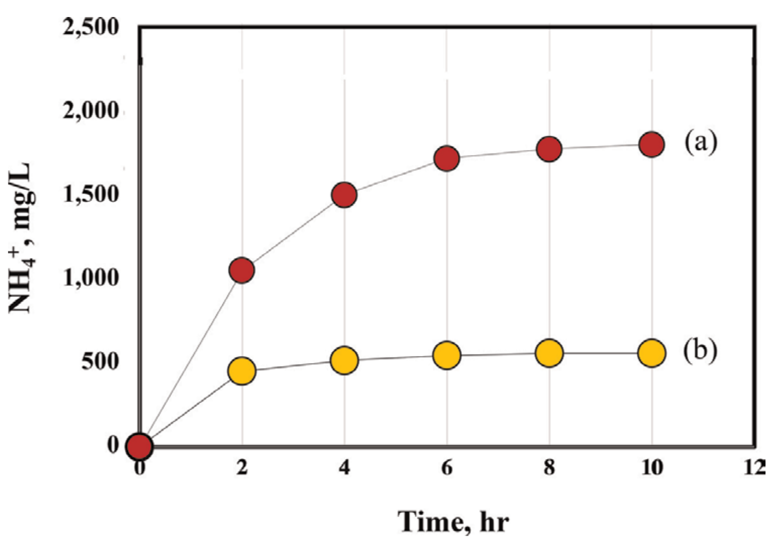

Figure 3.
The NH_4^+ concentrations in the recovery column (a) with and (b) without sulfuric acid.

NH_3 dissolution process. An earlier study has reported that the shape and the behavior of air bubbles also affect the gas solubility in water [22].

As **Figures 2** and **3** show, our process ensures a high recovery of NH_3 from manure digestate liquid by taking advantage of efficient chemical reactions, eqs. 3 and 5. The chemicals used are abundant and affordable. Still, the results shown in **Figures 2** and **3** are simply a proof of concept. The process can be improved by adjusting the kinetic parameters such as the mass transfer and the retention time of the air bubbles inside the columns. It should be noted, however, that eq. 3 produces sodium as a by-product which should be removed before spraying on croplands.

3.1.2 Highly concentrated NH_4 solution

The above operation was repeated by replacing the spent formulated sample a new one in the stripping column and dissolving the stripped NH_3 gas by reaction with sulfuric acid in the recovery column until the NH_4^+ concentration in the recovery column no longer increased. The NH_4^+ concentration in the recovery column is determined by the solubility of the product, $(NH_4)_2SO_4$, which is 744 g/L at ambient temperature. With this concentration, the concentration of NH_4^+ is 19%, a theoretical number. We reached 18% in our experiment. If nitric acid is used, the NH_4^+ concentration would be more than 33%, given the solubility of NH_4NO_3 in water, 1500 g/L.

To demonstrate the liquefaction of the highly concentrated NH_4^+ solution we prepared, we set up an experiment to produce liquefied NH_3. We heated 100 mL of the 18% NH_4^+ solution at 90°C, vaporizing NH_3, and sent the NH_3 gas, through a glass condenser into a metal cylinder half-submerged in iso-propanol which was cooled to −60°C by dry ice. The moisture generated by heating the N solution at 90°C was captured by a desiccant inside the condenser. At −60°C, the NH_3 gas is liquefied, while the other gases N_2 and O_2 stay as gas, being released to the atmosphere. About 50 mL of liquefied NH_3 was collected. The volume of the recovered liquid NH_3 was limited by the volume of the metal cylinder. Hence, no quantitative recovery rate was assessed. Yet, this demonstrates a possibility of NH_3 liquefaction from an 18% NH_4^+ solution. The technology for NH_3 liquefaction and the liquid NH_3 transportation infrastructure already exist. The same experiments should be repeated by using a real DS sample for validation.

Very few studies have been published to report the nitrogen concentration as high as 18% in recovering nitrogen from manure liquid. The high nitrogen concentration was made possible by dissolving the NH_3 gas into the water with a highly soluble acid. The other gases such as N_2, O_2, and CO_2 gases inside the air bubble can interfere with the NH_3 gas dissolving into the water. When the rising velocity of the bubbles is high, the NH_3 gas can be carried away by the other gases which are not water soluble except for CO_2 which dissolves somewhat. Using plastic packing materials inside the recovery column can help slow down the rising velocity.

As to the economic benefit, it is difficult to estimate since there is no market for renewable ammonia at this moment. Still, the price of N fertilizers has been going up significantly, due to the increase in the price of natural gas (NG), and can be unpredictable, given geopolitical reasons such as the economic sanction against Russia, a large exporter of N fertilizers. Using the recycled ammonia can help farmers save money. In addition, many large companies are investing in what is called Green Ammonia which uses water electrolysis followed by the Haber-Bosch process without using NG [23]. It is known that the production of Green Ammonia can cost four to five times as much as the conventional ammonia due to the high cost of water electrolysis [23]. Our process does not use electrolysis, nor does it produce NH_3.

It simply recovers NH_3 from the wastewater. Though a comprehensive cost-benefit comparison is not straightforward for Green Ammonia and our renewable NH_3, it should be clear that recovering NH_3 is much cheaper than producing it, given the extreme chemical stability of water and N_2, both of which are the raw materials for Green Ammonia. Both our renewable NH_3 and Green Ammonia should be qualified as non-fossil-based ammonia, and the demand for such ammonia is expected to grow massively high in the future [24].

3.2 Protein recovery

3.2.1 Composition of DS

Table 3 summarizes the compositions of the original DS sample and the leftover solid after the extraction of protein by THP on a dry matter basis. The condition for THP was the following: heating the DS sample at T_1 = 100°C for 1 hour followed by heating it further at T_2 = 160°C for 1 hour.

Almost 60% of the original protein was extracted by THP. Phosphorous mostly stayed in the leftover solid, while potassium dissolved in the solution after THP.

3.2.2 Protein recovery yield

We analyzed the protein recovery yield, $\eta_{recovery}^{protein}$, defined by the following equation:

$$\eta_{recovery}^{protein} = 100 \times \frac{[W_{hydrolysate}]}{[W_{protein}]} \tag{7}$$

where $[W_{hydrolysate}]$ and $[W_{protein}]$ refer to the weights of the protein hydrolysates (PHs) in the reaction solution after THP and the weight of the protein in the original sample prior to THP, respectively. The THP condition was the following: heating the DS sample at T_1 = 100°C for 1 hour followed by heating it further at T_2 = 160°C for 1 hour. **Table 4** summarizes the recovery yield under this condition, showing a reasonably high yield.

The numbers listed in **Table 4** were determined by AAA. The experiments were performed in triplicate and an error of $\eta_{recovery}^{protein}$ was within 3%. Vanotti et al. did not include the recovery yield in their patent [17].

The protein in manure digestate solid may be embedded in a complex solid matrix or trapped in a web of lignocellulosic components such as cellulose, hemicellulose, and

Solid sample	Protein[b]	P	K	Hemicellulose	Cellulose	Lignin	Others[c]
Before THP	37.2	2.1	1.5	9.3	18.2	30.2	1.5
After THP[d]	14.8	3.5	0.1	12.7	24.9	41.5	2.5

[a] *Dry matter basis.*
[b] *Obtained by multiplying the Kjeldahl nitrogen by 6.25. For the analytical method, refer to our earlier publication [20].*
[c] *Alkali metals such as Na, Ca, and Mg.*
[d] *The leftover solid after THP recovered by filtration by a screen with 90 μm mesh, dried in an oven overnight, and ground by a pestle for analysis.*

Table 3.
Compositions of DS samples before and the leftover solid after THP (wt.%)[a].

$W_{protein}$, g[a]	$W_{hydrolysate}$, g[a]	$\eta^{protein}_{recovery}$, %
9.21	5.55	60.26

[a]*Dry matter basis.*

Table 4.
Protein recovery yields by THP.

lignin. The interactions between the protein and the rest of the components in the solid may be hydrophobic in nature or electrostatic in nature associated with the functional groups of constituent amino acid residues of the protein. It is known that the dielectric constant of water decreases at high temperatures [25]. This creates two unusual characteristics for water: water favoring hydrophobic interactions and obstructing electrostatic interactions [25]. Hence, the first heating step of THP may interfere with the hydrophobic or the electrostatic interactions which may keep the protein trapped inside the solid matrix. It is also known that the pH of water goes down at high temperatures from around 7 [25]. Once the protein is released from the solid phase and dissolved into the solution phase, it should experience an acidic environment created by a low pH which may cause hydrolysis of the extracted protein. Accordingly, the dissolved protein may undergo hydrolysis, yielding short-chain peptides or individual amino acids. Although this is only speculation, the results show that protein can be extracted at a reasonably high yield by the two-step THP and what was extracted from DS was a mixture of oligopeptides and amino acids, as is shown below.

3.2.3 Molecular weight (MW) distributions

Figure 4 displays the SDS-PAGE band images for the PH prepared under the THP condition described above. The PH exhibited very few lines, indicating very few fractions within the range analyzed.

Figure 5 exhibits the MALDI-TOF mass spectra for the PH. The reference peptide, shown at 1046.79 m/z, was added to the sample prior to the measurements for the concentration of PH relative to the reference below the MW of 1000 Da. Contrary to the SDS-PAGE images, there are a number of peaks below 1000 Da. Peptides in this region were low-MW peptides such as oligopeptides or free amino acids. The concentration of the PH in **Figure 5** can be calculated from the peak positions and the intensities of each signal relative to the reference. It was about 3.9 g/L which is about 70% of $W_{hydrolysate}$ in **Table 4**.

Based on the results from SDS-PAGE and MALDI-TOF mass spectroscopy, we conclude that our PH had more low MW fractions than the higher MW fractions. Earlier studies reported low MW peptides exhibited antioxidant activities [26–28].

Next, we will subject our PH to antioxidant activities.

3.2.4 ORAC against the peroxyl and hydroxyl radicals

Figure 6a and **b** show the inhibition of the peroxyl radical attack against fluorescein protein by Trolox and PH, respectively, as a function of the logarithm of the sample concentration, *C*. This assay is important since lipid molecules constituting cell membranes are prone to become peroxyl radicals that attack DNA, protein, and other molecules in a cell [29]. The curve profile shown in **Figure 6b** is very similar to that in **Figure 6a**. In fact, the values of IC_{50} for PH and Trolox were very close: 7.67 and

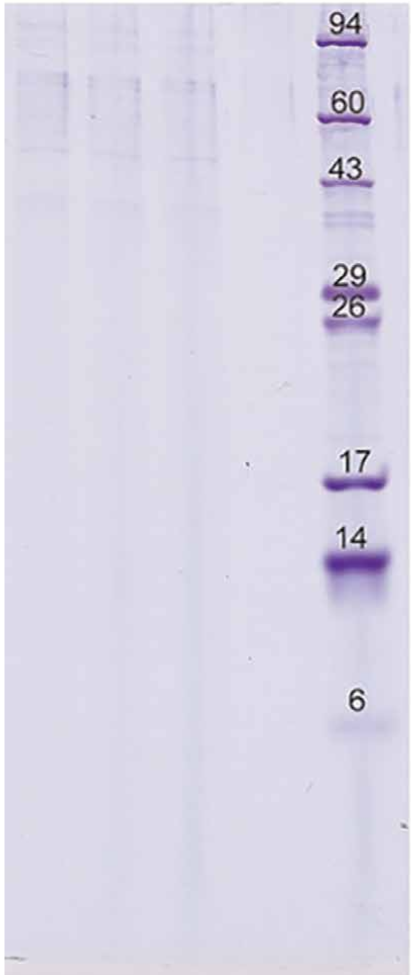

Figure 4.
SDS-PAGE image of PH extracted from DS. The measurements were performed in triplicate. The numbers on the right side are the MW markers in kDa.

8.08 mg/L, respectively. This observation demonstrates that the antioxidant activity of PH was as strong as Trolox against the peroxyl radicals. IC_{50} refers to the concentration of the sample at which the inhibition is 50%. The experimental error for IC_{50} was within 1 mg/L which was estimated over triplicate experiments.

Figure 7a and **b** display the inhibition of the hydroxyl radical attack against fluorescein protein by Trolox and our PH, respectively, as a function of the logarithm of *C*. Hydroxyl radicals are often generated inside a cell in the presence of metal ions such as Fe(II), Cu(I), and Co(II) and attack organic molecules involved in metabolic reaction pathways [30]. We observed a significant difference between the two samples: the inhibition by our PH reached 100% when log *C* was 1.5, while Trolox did not reach 100% inhibition even when log *C* was 2. IC_{50} of 107.6 mg/L for our PH was less than 1/7 of that of Trolox, 741 mg/L. The values of IC_{50} imply that the antioxidant activity of our PH was more than seven times as strong as Trolox. The strong antioxidant activities of our PH are consistent with the previous studies on peptides

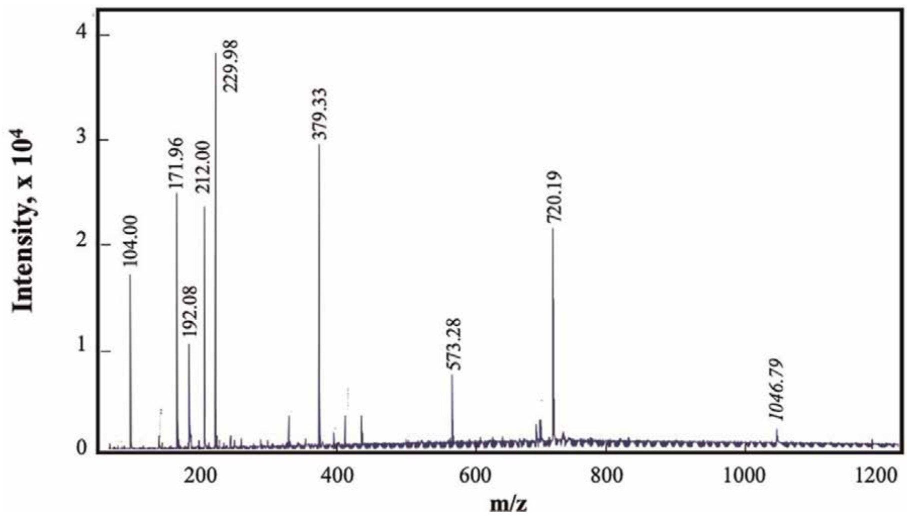

Figure 5.
MALDI-TOF-mass spectrum of PH. A signal for a peptide with a known MW is included as a reference at 1046.79 m/z.

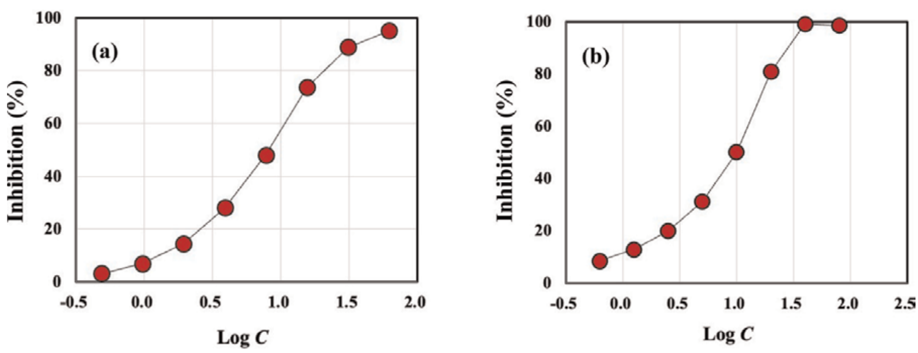

Figure 6.
The inhibition of the peroxyl radical attack against fluorescein protein by (a) Trolox and (b) PH, respectively, as a function of the logarithm of the sample concentration, C.

[26–28, 31–40]. A theoretical study on the antioxidant activity of peptides has been published [41].

The DS sample included some non-protein nitrogen compounds which were not removed from the PH sample prior to the ORAC assay; therefore, their contributions to the inhibition of the radicals cannot be ignored. Our data only demonstrates that the extracted compounds from the DS sample by our THP and recovered by UF with a 150 kDa membrane inhibited both peroxyl and hydroxyl radicals to the extent that the ability to inhibit the former radical was comparable to that of Trolox and the ability to inhibit the latter was seven times stronger than Trolox.

Our assay is an in-vitro test, and the results should be considered preliminary. Further study is warranted to confirm the antioxidant activity of our PH. If confirmed, the recovered protein hydrolysate could be sold as antioxidant feed additives. Currently, a 50% feed-grade vitamin E is sold at ∼$13.5/kg [42]. Using the numbers in **Table 2**, the volume of the recovered protein from manure digestate solid generated

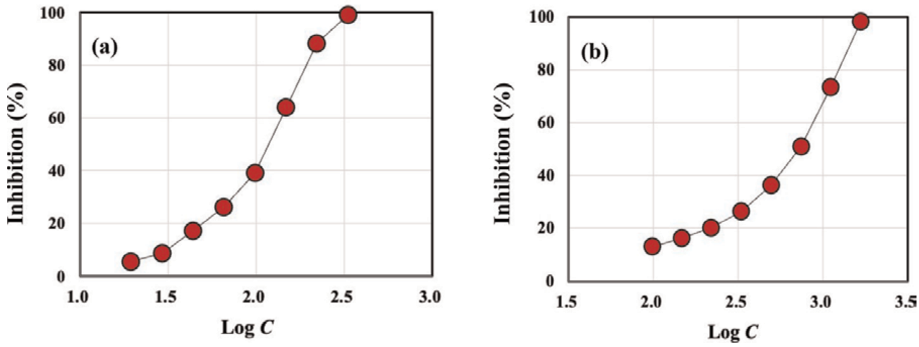

Figure 7.
The inhibition of the hydroxyl radical attack against fluorescein protein by (a) Trolox and (b) PH, respectively, as a function of the logarithm of C.

on a dairy farm would be about 5000 tons/year at the recovery yield of 60%. This would provide an annual revenue of $137 million/year which could potentially overshadow the revenue from selling the milk. It should be noted that this is only a rough estimate under a hypothetical scenario.

The GHG emissions from agricultural activities in the U.S. were 641 Mt of CO_{2eq} in 2019 [43]. Of that volume, 58% was due to the N_2O emission caused by spraying nitrogen fertilizers including manure/digestate liquids through the mechanism mentioned earlier, and 13% was primarily due to the N_2O emissions by manure management mostly from manure storage such as lagoons. N_2O generated by manure management is mainly produced from organic nitrogen, mostly protein, in manure. By recovering protein from manure before manure management through the protein recovery process described in this chapter, we could potentially reduce about 83 Mt of CO_{2eq} emissions, assuming the above 13% was generated by decomposition of protein in manure. 30 to 50% of the N_2O emissions caused by spraying N fertilizers and manure/digestate liquids originate from applications of animal manure which includes organic nitrogen [3, 4]. Hence, practicing a combination of the protein and NH_3 recovery processes described in this chapter could potentially reduce about 109–186 Mt of CO_{2eq} emissions. Altogether, up to 269 Mt of CO_{2eq} emissions could be removed by the combination of the two processes. This number is about 42% of the total GHG emissions from agricultural activities in the U.S. To evaluate these estimates, we assumed 100% protein and NH_3 recovery rates by both recovery processes. Though the actual number will be lower, a significant volume of GHG emissions can be still reduced by the two processes. The potential reduction of eutrophication caused by nitrogen runoff cannot be ignored through our processes.

4. Conclusion

The two processes described here, the NH_3 and the protein recovery process from manure digestate, have shown the high recovery rates. Our processes can be directly applied to the current practice of the solid-liquid separation of manure digestate from which both solid and liquid are otherwise sprayed on lands. The protein recovery process can be applied to the solid, while the NH_3 recovery process can recover NH_3 from the liquid, with the solid and liquid coming from ADs which are rapidly being adopted by livestock farmers. Using the two processes can significantly limit the leakage of N from the digestate into the environment.

Furthermore, our two processes can produce value-added products including protein-based antioxidant feed additives and concentrated N solutions from which renewable nitrogen fertilizers or non-zero carbon renewable energy source can be recovered. These products can help close the nitrogen loop in the livestock operation that is currently broken, given abundant applications of synthetic fertilizers and frequent use of protein-based feeds. The nitrogen in the recovered protein from manure/digestate solids can go back to animals as feed, while the nitrogen recovered from manure/digestate liquids can grow crops, and the nitrogen in the crops can be recycled back to animals as feed as well. Additionally, our recovered NH_3 is renewable NH_3 produced without fossil fuels and highly energy-intensive processes such as the Haber-Bosch process or water electrolysis.

Intense animal operations (IAO) are expected to grow, given the increasing demand for meats and dairy products worldwide. Accordingly, regulations on manure management will be likely tightened to keep the environmental consequences by IAO under control. Yet, regulations can go only so far as to mitigate the environmental consequences. Our processes add economic incentives to livestock farmers by bringing extra revenue streams which will help livestock farmers, some of whom may be under financial stress due to increasingly higher costs for the operations.

Authors' contribution statements

ANT contributed by preparing samples and conducting THP experiments; KT contributed by the conceptualization of experiments, manuscript preparation, data analysis, and review editing. All authors have read and agreed to the manuscript to be submitted.

Author details

Abigail N. Tasaki[1] and Ken Tasaki[2,3]*

1 South High School, Torrance, CA, United States

2 Tomorrow Water, Anaheim, CA, United States

3 Palos Verdes Research, Redondo Beach, CA, United States

*Address all correspondence to: kt@bkt21.com

References

[1] OECD-FAO Agricultural Outlook 2020–2029. Chapter 6: OECD and Food and Agriculture Organization of the United Nations. 16 July 2020. pp. 163-177

[2] Bouwman L, Goldewijk KK, Van der Hoek KW, Beusen AH, Vuuren DP, Willems J, et al. Exploring global changes in nitrogen and phosphorus cycles in agriculture induced by livestock production over the 1900–2050 period. Special Features - PNAS. 2013;**110**: 20882-20887. DOI: 10.1073/pnas.1012878108

[3] Davidson EA. The contribution of manure and fertilizer nitrogen to atmospheric nitrous oxide since 1860. Nature Geoscience. 2009;**2**:659-662

[4] Oenema ON, Wrage GL, Velthof JW, van Groenigen J, Kuikman PJ. Trends in global nitrous oxide emissions from animal production systems. Nutrient Cycling in Agroecosystems. 2005;**72**: 51-65

[5] Chadwick DR, Pain BF, Brookman SKE. Nitrous oxide and methane emissions following application of animal manures to grassland. Journal of Environmental Quality. 2000;**29**: 277-287

[6] Petersen SO, Nissen HH, Lund I, Ambus P. Redistribution of slurry components as influenced by injection method, soil, and slurry properties. Journal of Environmental Quality. 2003; **32**:2399-2409

[7] Fertilizer Use and Price. USDA, Economic Research Service. 2019

[8] U.S. Department of Agriculture. 2007 Census of Agriculture: United States Summary and State Data. Vol. 1. AC-07-A-5. 2009

[9] Vaneeckhaute C, Lebuf V, Michels E, Belia E, Vanrolleghem PA, Tack FMG, et al. Nutrient recovery from digestate: Systematic technology review and product classification. Waste and Biomass Valorization. 2017;**8**:21-40

[10] Starmans DAJ, Timmerman M. Apparatus for the ammonium recovery from liquid animal manure. Applied Engineering in Agriculture. 2013;**29**(5): 761-767. DOI: 10.13031/aea.29.10018

[11] Pedros PB, Simon MM, Brown SH. Biocast Process. U.S. Patent 8,398,855 B1. 2013

[12] Riaño B, Molinuevo-Salces B, Vanotti MB, García-González MCG. Application of gas-permeable membranes for-semi-continuous ammonia recovery from swine manure. Environments. 2019;**6**:32. DOI: 10.3390/environments6030032

[13] Chen S, Liao W, Liu C, Wen Z, Kincaid RL, Harrison JH, Elliott DC, Brown MD, Solana AE, Stevens DJ. Value-Added Chemicals from Animal Manure. PNNL-14495. Richmond, Washington: Pacific Northwest National Laboratory; 2003

[14] The Midwest Plan Service. Livestock Waste Facilities Handbook. Ames, IA: Iowa State University; 1993

[15] Robinson TP, William GR, Conchedda GW, Van Boeckel TP, Ercoli V, Palamara E, et al. Mapping the global distribution of livestock. PLOS ONE. 2014;**9**(5):e96084

[16] Kim R, Zinbo M, Adams JA, Young WC. Nonbiodegradable organic compounds found in automotive spraybooth scrubber water. Water Environment Research. 2000;**72**:405-412

[17] Vanotti M, Szogi A. Extraction of amino acids and phosphorus from biological materials. U.S. Patent Application 20190071370. 7 March 2019

[18] Nastića N, Švarc-Gajića J, Delerue-Matosb C, Barroso MF, Soares C, Manuela MM, et al. Subcritical water extraction as an environmentally-friendly technique to recover bioactive compounds from traditional Serbian medicinal plants. Industrial Crops and Products. 2018;**111**: 579-589

[19] Liang X, Fan Q. Application of sub-critical water extraction in pharmaceutical industry. Journal of Materials Science and Chemical Engineering. 2013;**1**:1-6. DOI: 10.4236/msce.2013.15001

[20] Hasan MH, Mahlia TMI, Mofijur M, Rizwanul FIM, Handayani F, Ong HC, et al. A comprehensive review on the recent development of ammonia as a renewable energy carrier. Energies. 2021;**14**:3732

[21] Tasaki K. Chemical-free recovery of protein from livestock manure digestate solid by thermal hydrolysis. Bioresources and Bioprocessing. 2021;**8**:60

[22] Li X, Wang W, Zhang P, Li J, Chen G. Interactions between gas–liquid mass transfer and bubble behaviours. Royal Society Open Science. 2019;**6**:190136. DOI: 10.1098/rsos.190136

[23] Tullo AH, editor. Is ammonia the fuel of the future? Chemical and Engineering News. 2021;**2021**:99

[24] Patonia A, Poudineh R. Ammonia as a Storage Solution for Future Decarbonized Energy Systems. Oxford, UK: The Oxford Institute for Energy Studies; 2020

[25] Plaza M, Turner C. Pressurized hot water extraction of bioactives. Trends in Analytical Chemistry. 2017;**71**: 39-54

[26] Wu HC, Shiau CY, Chen HM, Chiou TK. Antioxidant activities of carnosine, anserine, some free amino acids and their combination. Journal of Food and Drug Analysis. 2003;**11**: 148-153

[27] Adhikari B, Dhungana SK, Ali MW, Adhikari A, Kim ID, Shin DH. Antioxidant activities, polyphenol, flavonoid, and amino acid contents in peanut shell. Journal of the Saudi Society of Agricultural Sciences. 2019;**18**: 437-442

[28] Liu R, Xing L, Fu Q, Zhou GH, Zhang WG. A review of antioxidant peptides derived from meat muscle and by-products. Antioxidants. 2016;**5**:32. DOI: 10.3390/antiox5030032

[29] Kehrer JP, Robertson JD, Smith CV. Free radicals and reactive oxygen species. In: McQueen CA, editor. Comprehensive Toxicology. 2nd ed. Amsterdam: Elsevier; 2010. pp. 277-307. DOI: 10.1016/B978-0-08-046884-6.00114-7

[30] Levine JS. Biomass burning: The cycling of gases and particulates from the biosphere to the atmosphere. In: Holland HD, Turekian KK, editors. Treatise on Geochemistry. 2nd ed. Elsevier; 2014. pp. 139-150

[31] Feng P, Ding H, Lin H, Chen W. The antioxidant protein database. Scientific Reports. 2017;7:7449. DOI: 10.1038/s41598-017-08115-6

[32] Ye N, Hu P, Xu S, Chen M, Wang S, Hong J, et al. Preparation and characterization of antioxidant peptides

from carrot seed protein. Journal of Food Quality. 2018:8579094

[33] Kim JM, Liceaga AM, Yoon KY. Purification and identification of an antioxidant peptide from perilla seed (*Perilla frutescens*) meal protein hydrolysate. Food Science & Nutrition. 2019;**7**:1645-1655. DOI: 10.1002/fsn3.998

[34] Yang J, Hu L, Cai T, Chen Q, Ma Q, Yang J, et al. Purification and identification of two novel antioxidant peptides from perilla (*Perilla frutescens* L. Britton) seed protein hydrolysates. PLoS One. 2018;**13**(7):e0200021

[35] Davalos A, Miguel M, Bartolome B, Lopez-Fandino R. Antioxidant activity of peptides derived from egg white proteins by enzymatic hydrolysis. Journal of Food Protection. 2004;**67**: 1939-1944

[36] Sila A, Bougatef A. Antioxidant peptides from marine by-products: Isolation, identification and application in food systems. A review. Journal of Functional Foods. 2016;**21**:10-26

[37] Wang J, Liao W, Nimalaratnel C, Chakrabarti S, Wu J. Purification and characterization of antioxidant peptides from cooked eggs using a dynamic in vitro gastrointestinal model in vascular smooth muscle A7r5 cells. npj Science of Food. 2018;**2**:7. DOI: 10.1038/s41538-018-0015-7

[38] Zou TB, He TP, Li HB, Tang HW, Xia EQ. The structure-activity relationship of the antioxidant peptides from natural proteins. Molecules. 2016;**21**:72. DOI: 10.3390/molecules21010072

[39] Hook VYH, Burton D, Yasothornsrikul S, Hastings RH, Deftos LJ. Proteolysis of ProPTHrP (1–141) by prohormone thiol protease at multibasic residues generates PTHrP-related peptides: Implications for PTHrP peptide production in lung cancer cells. Biochemical and Biophysical Research Communications. 2001;**285**:932-938

[40] Qian ZJ, Jung WK, Byun HG, Kim SK. Protective effect of an antioxidative peptide purified from gastrointestinal digests of oyster *Crassostrea gigas* against free radical induced DNA damage. Bioresource Technology. 2008;**99**:3365-3371

[41] Leung R, Venus C, Zeng T, Tsopmo A. Structure-function relationships of hydroxyl radical scavenging and chromium-VI reducing cysteine-tripeptides derived from rye secalin. Food Chemistry. 2018;**254**: 165-169

[42] Available from: https://www.echemi.com/produce/pr2203314355-vitamin-e-50-feed-grade-50-snc-good-fortune.html

[43] Massey R, Klentzy D. Agriculture and Greenhouse Gas Emissions. University of Missouri Extension; 2021

Chapter 8

Recent Advances and Application of Biotechnology in the Dairy Processing Industry: A Review

Muhammed Nurye Gebeyehu

Abstract

Worldwide continuous demand for milk and milk products triggers different stakeholders in the dairy sector, which leads to the establishment of modern dairy farms, processing companies, and the invention of artificial intelligence. This dramatic change in the sector boosted milk production and reduce the impact of dairying on the environment. There has been a tremendous improvement in all unit activities such as separation, standardization, pasteurization, homogenization, and packing due to modern innovations in the fluid milk processing business. Advanced technologies in milk processing are applied to extend the shelf life, enhance the nutritional quality and safety of dairy products and health advantages without altering its physicochemical characteristics. Generally, the use of recent technologies in milk production has a significant impact to address the demand for milk and milk products, poverty alleviation, reducing GHG emissions, and other global challenges. The dairy industry can benefit greatly from new advancements and innovations in modern biotechnology, such as rDNA technology, transgenics, probiotics, bio preservatives, recombinant enzymes, starter culture, and improved bioprocess engineering tools by producing novel foods customized for specific consumers. While biotechnology brings incredible benefits, it also persuades a potential impact on health and the environment. Therefore, its application needs great intention.

Keywords: biotechnology, dairy product, milk, technology, processing

1. Introduction

While milk production per cow has risen dramatically in the last three decades through research that emphasizes efficient and sustainable milk production alongside productivity improvement, farmers still seek the wisdom of attempting to increase milk production with long production years in order to alleviate their fear of dairy cows' short productive lifespan. Moreover, achieving maximum productivity using scarce natural resources is the greatest challenge for dairy farmers, which can be addressed by implementing dairy farming innovations on every farmer's farm. The application of innovations at all stages of production, from fodder cultivation to milk marketing, is a critical need in today's world [1]. As a result, scientists and researchers devise a variety of innovations to increase resource efficiency and/or provide information to farmers. In the last two decades computerized or automated technologies, such as computerized feed delivery and milking systems, on-farm computers to manage dairy records, automatic take-offs for milking units, and a holding pen with an udder washer that uses computer hardware and software, have been discovered. Moreover, Pulsed electric fields, high hydrostatic pressure, high-pressure homogenization, ohmic and microwave heating, microfiltration, pulsed light, UV light processing, and carbon dioxide processing are some of the emerging dairy processing technologies that could reduce energy consumption and greenhouse gas emissions [2]. The efficiency of processing technologies is also enhanced by using bacteriocins. Furthermore, milk processing has a significant impact on small-scale dairy producers to generate higher cash incomes than selling raw milk and offers better opportunities to reach regional and urban markets. Milk processing can also help to deal with seasonal fluctuations in milk supply [3].

Despite the fact that achieving expected animal productivity at a lower cost for greater economic returns is dependent on the ability to transfer these innovations from the lab to the field in the dairy farming system, innovations have a significant impact in overcoming the sector's current challenges. Consequently, to raise milk production with minimal GHG emissions, improved animal husbandry techniques should be adopted in smallholder household dairy farms [1] along with technological innovations.

Biotechnology's application in the dairy industry has a substantial role that improve milk production, animal health, and food processing. Genetically modified microorganisms such as bacteriocins and probiotics have been associated with lowering the risks of type 2 diabetes, metabolic syndrome, and heart disease, as well as better weight management [4]. Besides, probiotic products contain microorganisms that are viable, specific, and effective in critical nutritional physiology systems. *Lactobacillus, Bifidobacteria, Saccharomyces,* and *Streptococcus* are some of the bacteria found in fermented dairy products [5, 6]. Bio-preservation with natural preservatives has a positive effect on consumer health promotion in addition to improving food safety and shelf-life as a technological effect [4].

Furthermore, the application of technology has had a significant impact on addressing several challenges faced by the food industry on food safety, preservation, nutrition, and allergies, food security, changing consumer needs, environmental concerns, economic viability, and policy reforms. Dairy processing companies around the world need efficient, low-cost, and automated innovations to increase profits and meet the demand of a wide range of products. Therefore the aim of this paper is to recapitulate and combine the different information on current development in milk production and processing.

2. Overview of the global dairy industry

Global milk production trends increases dramatically in the last 30 years from 530 million tonnes in 1988 to 843 million tonnes in 2018 [3]. Over the next ten years, global milk production is expected to increase at a rate of 1.6 percent each year (to 997 Mt. by 2029), outpacing most other major agricultural commodities [7], however, the expected growth remains tight in 2022, with only a 0.6 percent increase projected and EU countries produced below the expectation, especially New Zealand and Australia [8]. Moreover, the global milk production reported in January 2022 was lower by 1.6%

than the previous year's counterpart [9]. USDA is estimating just a 0.7% rise in milk production in 2022, which is bullish for milk prices.

On the other hand, milk production growth is predicted to decline by 0.5 percent per year in the European Union (EU) and reach 162 million tonnes by 2031. However, organic milk production in this region is predicted to increase (to 8% by 2031), resulting in economic gains, environmental benefits, and improved animal welfare [10]. The growth of average global milk production is achieved by improving the dairy cow production performance than the number of herds. Besides the large volume of milk consumed in the form of fresh dairy products, including pasteurized and fermented products, due to a significant increase in milk product demand in developing nations, the proportion of worldwide fresh dairy products consumption is predicted to rise over the next decade OECD [11]. In developed countries, processed dairy products are preferred, whereas, in underdeveloped countries, fresh dairy products account for more than 75% of the average per capita daily intake of milk solids. In underdeveloped countries, regional differences are enormous, with fresh dairy product consumption ranging from 99% in Ethiopia to 5.8% in the Philippines OECD [11].

Furthermore, according to Minj *et al*. [12], the production, storage, and distribution of various types of dairy products, as well as the management of dairy-related data, are significantly influenced by global milk production and processing. According to literature evidence, various processing technologies have been used to produce several types of milk and milk products. Market milk, flavored milk, cream, butter, butter oil/ghee, condensed and evaporated milk, milk powder, fermented milk, yogurt, cheese, ice cream, and indigenous dairy products are among the most common processed dairy products. The major processing technologies necessitate a robust setup for continuous production and maintaining final product quality [12].

2.1 Revolutions in milk production and milking

Manual or hand milking is a time-consuming, labor-intensive process and the milk preserving process is also unsanitary, moreover, a bacterial infection in milk can occur as a result of a manual process. Wondatir [13] proposed a novel robotic milking technology that can grab the milking claws of cows. Introducing auto-milking has solved this problem more efficiently through lowering costs and manpower, automatically preserving the milk by using various smart cooling tanks. Few models of low-cost, nonelectric milking machines are also developed considering the locality and need of dairy farms. Milk that did not fit for human consumption is diverted to a separate container. The sensors in the automatic milking machine play important role in detecting the readiness of teats for milking and also identify impurities, color, and quality of milk. The majority of auto-milking systems rely on two components: a computer and specialized herd management software. This material can perform activities such as collecting the animal, cleaning the animal before milking, attaching the milking equipment, extracting milk, removing the equipment, and routing the animal out of the special area Muhammad Osama [14]. The robotic feeding system can calculate the effectiveness of feed for milk production as well as a cow's nutritional requirements.

The invention and introduction of highly efficient automatic milk meters that built into robotic milking systems of cows are extremely important to control the main technological parameters in the process of milking (live weight, temperature, electrical conductivity, and so on) [15, 16]. Additionally, it is also a remarkable role to determine different stages of lactation, heat periods, somatic cell count, motor activity, and other zootechnical registration parameters. Viguier et al. [17] revealed the use of SCC as an alternative method of detecting mastitis. Hereafter numerous sensors are used in the production of high-quality milk and the use of microchip technologies has resulted in faster results. Furthermore, with these technologies, you will be able to diagnose mastitis more successfully with more effective tests and results with a wider angle and more accurate results. Milk conductivity and milk appearance are commonly used on farms. Other methods, on the other hand, provide another early mastitis detection for a quick and accurate decision to cure the disease.

Moreover, an automatic milking system has a priceless impact not only on milking but also on management systems including feeding, cow traffic, cow behavior, grazing, milk quality, and animal health by using electronic devices or sensors. A large number of research studies have reported analyses of AMS impact on specific aspects such as milk yield/quality [18, 19], animal behavior/health/welfare herd management [20] performance, and labor efficiency [21, 22]. Some studies reported a 2 to 12 percent increase in milk production in cows milked 2+ times per day in AMS compared to cows milked twice per day in traditional milking parlors. However, the result of Hansen et al. [23] revealed that the use of AMS did not show an increase in milk production, especially for prim parous cows [24–26].

The machine can monitor the state of herd productivity and express milk analysis is a critical condition for effective milk production and industry competitiveness. Express analysis of each animal's milk allows you to assess the animal's health and productivity with confidence. The data obtained from the automated machine will be processed and used to control the level of productivity and identify problems for immediate corrective action [27].

The first commercial AMS in dairy farms was introduced in the Netherlands in the 1990s, and about 50,000 units were adopted worldwide by 2020 [28]. AMS is mainly concentrated in Europe (90%), Canada (9%), and other countries (1%). However, it is expected that by 2025, 50% of dairy cows in North-Western Europe will be equipped with AMS [29].

To recapitulate, the innovation of robotic milking machines is useful in eliminating the pressure on human labor and maintaining a hygienic milking process with remarkable improvement in milk production, as well as managing every aspect of management and reproduction in the farm by incorporating an automated milk meter on it.

2.2 Recent advances in milk processing or dairy industry

In the last two decades, major technological advances in the fluid milk processing industry have been observed, with a significant improvement in all unit operations such as separation, standardization, pasteurization, homogenization, and packaging. Besides, many advances have been made in terms of production capacity, automation, and sanitary operation [30]. Traditionally, milk is processed by heating it to a specific temperature for a fixed period of time, which results in a significant reduction in the microbial population [31]. However, recently developed nonthermal processing methods are ideal for milk and other food staff with higher performance of eliminating microorganisms or any other biological entities without causing a significant temperature rise, thereby preventing a chain of undesirable reactions in foods [31]. High-pressure processing (HPP), microfiltration, centrifugation, pulsed electric field (PEF), ultraviolet light (UV), and cold plasma processing are among the widely used nonthermal processing. Moreover, automated technologies have also been developed to reduce labor costs and losses during processing, including automated clean-in-place (CIP) system, inclined film scraped surface heat exchanger (ISSHE), automated spray dryer, membrane processing (ultra filtration (UF), reverse osmosis (RO), micro-filtration (MF), nano filtration (NF), and electrodialysis). Ultrasonic processing or sonication is a promising alternative technology in the food industry as it has the potential to improve the technological and functional properties of milk and dairy products. Furthermore, High-intensity ultrasound (HIU) is a promising emerging technology, specially designed for economy, simplicity, and energy efficiency. HIU has multiple benefits either in the processing or evaluation of products [32]. It also offers a great potential to control, improve, and accelerate processes without damaging the quality of food and other products.

2.2.1 High-pressure processing (HPP)

A nonthermal method of food and dairy product preservation and sterilization in which a product is subjected to extremely high pressure, causing some microbes and enzymes in the food to be inactivated [33]. Research on raw milk treated with high pressure has shown that HPP treatment produces raw milk of comparable quality to pasteurized milk, as it is equally successful in eradicating pathogenic and spoilage microorganisms. When compared to foods with a higher pH, such as milk, HPP proved effective at inactivating bacteria in both high- and low-acid food systems. It may influence the qualities of treated milk by modifying the fundamental features of milk ingredients [34]. In the food and dairy sector, high-pressure processing is a unique alternative to thermal processing [33] that includes a treatment chamber, a pressure generating system, a pressure transmission medium, and a pressure intensifier [34]. HPP was performed at 680 MPa for 10 minutes at room temperature and the number of microorganisms was reduced by 5–6 log cycles.

2.2.2 Pulsed electric fields (PEF)

The fundamental principle of PEF technology is the use of short pulses of high electric fields with durations ranging from microseconds to milliseconds and intensities ranging from 10 to 80 kV/cm. Short pulses (1–10 μs) generated by a high voltage (5–20 kV) pulse generator have been used to treat biological material or food placed between two electrodes installed 0.1–1.0 cm apart in a treatment chamber separated by an insulator [35].

The processing time is calculated by multiplying the number of pulse times with effective pulse duration. The applied high voltage results in an electric field that causes microbial inactivation. When an electrical field is applied, electrical current flows into the liquid food and is transferred to each point in the liquid because of the charged molecules present ([36]; as cited by [33]). PEF treatment has achieved a reduction in the microflora of milk with a shelf life similar to that of high temperature, short time (HTST) pasteurized milk.

The ability to control the amount of ohmic heating in food preservation (low-temperature processing) is the main benefit of PEF technology in liquid food pasteurization. This avoids the Maillard reaction, which affects the functional properties of food such as color, taste, and smell [37]. PEF is also effective in the inactivation of microorganisms such as *Salmonella typhimurium*, *Listeria innocua*, and *E. coli* up to 5.0 log cycles [38].

The method is highly scalable and can be incorporated into existing food processing lines. In comparison to traditional heat pasteurization technology, it is more energy-efficient [39]. Furthermore, PEF treatment chambers can be easily adapted to existing continuous-flow production lines for liquid food pasteurization [40]; however, achieving a homogeneous treatment may be an issue [41]. The main disadvantage of PEF technology is its effectiveness and efficiency, which are largely dependent on the liquid conductivity and viscosity [42].

2.2.3 Ultra-sonication

It refers to the application of sound waves at the frequency ($>$ 16 kHz) greater than the upper limit of human hearing through liquid, solid, or gases, which causes the formation of small bubbles (known as cavitation). While droplets reach the required size range, they collapse under near-adiabatic conditions, resulting in significant conditions both within the droplets and in the surrounding liquid that include intense shear forces, turbulence, and micro streaming effects. These ultrasound-induced physical effects are increasingly being used in food and dairy processing industries, its application is used to enhance whey ultrafiltration, extraction of functional foods, reduction of product viscosity, homogenization of milk fat globules, crystallization of ice and lactose, and the cutting of cheese blocks [33].

The use of ultrasound in traditional dairy processes has the potential to bring significant cost savings and improved product qualities to the dairy sector. Furthermore, as compared to other new technologies, the use of ultrasound as a processing technique has been deemed safe [43]. These technologies include low- and high-intensity ultrasounds. Low-intensity ultrasounds have been used to determine, evaluate, and define the physical features of foods, whilst high-intensity ultrasounds have been utilized to speed up specific biological, physical, and chemical processes during the handling and transformation of food products [44].

2.2.4 Cold plasma (CP)

Cold Plasma is an electrically powered gaseous state consisting of charged particles, free radicals, and some radiation that is known as the fourth state of matter. An electrical discharge [45] produces a partially or totally ionized plasma made up of photons (basically), ions, free electrons, and atoms in their fundamental or excited states. These species are classified as either “light” (photons and electrons) or “heavy” (remaining constituents) [46, 47].

Currently, CP undergoing extensive testing for the preservation of perishable commodities such as milk and milk products. The use of cold plasma (CP) techniques to preserve milk and milk products has been pushed as a revolutionary nonthermal technology. CP not only preserves the nutritional value of the food but also inactivates germs, eliminating the risk of resistance. Cold plasma was also discovered to disrupt enzymes involved in browning (color change) processes and the production of an off-flavor [48].

2.3 Membrane separation technology

It is a method of separating a liquid into two streams using a semipermeable membrane. The two streams are called retentate and permeate, respectively. Specific components of milk and whey can be separated using membranes with different pore sizes. Membrane filtering technology offers a variety of applications in the cheese industry, including boosting nutritional quality, improving compositional control and production by increasing total solid content, using whey during cheese manufacturing, and minimizing the need for rennet and starter culture. Concentrating milk before manufacturing cheese opens up a new market for the cheese industry, lowering costs and speeding up the entire process [49]. Membranes in the cheese industry concentrate the cheese milk, increasing yield and quality while controlling whey volume. It is now possible to recover growth factors from whey because of advancements in membrane technology [50]. Membrane filtration can basically be divided into four main technologies, which are as follows:

2.3.1 Microfiltration (MF)

Microfiltration is a membrane filtration technique that uses a membrane with an open structure and is powered by low pressure. The membrane allows dissolved components to pass while rejecting the majority of non-dissolved components. Microfiltration is widely used in the dairy industry to reduce bacteria and spores, remove fat from milk and whey, and standardize protein and casein.

2.3.2 Ultrafiltration (UF)

Ultrafiltration is a membrane filtration process that operates at medium pressure. Ultrafiltration works by passing most dissolved and non-dissolved components through a membrane with a medium open structure while rejecting larger components. UF is widely used in the dairy industry for whey protein concentration and milk protein concentration as well as standardization.

2.3.3 Nano-filtration (NF)

Nano-filtration is an intermediate step in the high-pressure membrane filtration process. In general, nano-filtration is a type of reverse osmosis in which the membrane has a slightly more open structure that allows primarily monovalent ions to pass through. The membrane rejects divalent ions to a large extent. Nano-filtration is primarily used in the dairy industry for specialized applications such as partial demineralization of whey, lactose-free milk, and whey volume reduction.

2.3.4 Reverse osmosis (RO)

Reverse osmosis is a high-pressure, membrane-based filtration process that uses a very dense membrane. In theory, only water passes through the membrane layer. Reverse osmosis is commonly used in the dairy industry for milk and whey concentration or volume reduction, milk solids recovery, and water reclamation.

2.4 Application of biotechnology in dairy processing

Recent biotechnological breakthroughs have emerged as a significant tool for developing quality features in livestock products, such as dairy and dairy-based products. In most developing nations, biotechnology has been used to improve food processing by using microbial inoculants to improve qualities such as flavor, scent, shelf life, consistency, and nutritional content of meals and dairy products. Probiotic food products are a rapidly expanding segment of functional food that has been well received by consumers. The food sector, on the other hand, is striving to offer a variety of probiotic foods other than dairy products with potential health benefits [51].

Moreover, modern biotechnology has brought up new and exciting opportunities in the dairy industry, making milk and milk products more accessible to the poor and meeting the demands of a larger population. Since the dairy industry's primary responsibility is to provide consumers with high-quality, nutritional, and affordable dairy meals, biotechnological intervention at various stages of milk production and processing has become a foregone conclusion [52]. It has provided us with delicious, nutritious, wholesome, handy, shelf-stable, and safe foods. As long as research and development efforts continue, biotechnology will inevitably have a greater impact on the food we eat. It has enormous potential for expanding the variety and quality of food available to humans, especially more healthy and appealing foods. It also appears likely that it will continue to provide benefits to food processing and safety monitoring as new technologies develop at a faster rate. Furthermore, the biotechnological application has a remarkable role in dairy product bio-preservation, probiotics manipulation, and production; enzyme production; milk derived bioactive peptides and other functional ingredients; and starter cultures technology and genetic manipulation.

2.4.1 Bio-preservation

Although recent advances in innovative modern technologies implemented in food processing and more stringent microbiological food-safety standards have reduced the incidences of foodborne illnesses and product spoilage, they do not completely eliminate the possibility of health risks associated with such foods. As a result, the food industry is always exploring novel techniques and methods to produce minimally processed, ready-to-eat food that retains its nutritional value, taste, and flavor. Bio-preservation like bacteriocin is an ideal choice to preserve ready-to-eat processed foods without altering their nutritional and chemical properties.

Bacteriocins are antimicrobial peptides that are deemed harmless since they are easily destroyed by mammalian gastrointestinal proteolytic enzymes. Furthermore, the majority of bacteriocin producers belong to lactic acid bacteria (LAB). Bacteriocins, whether purified or secreted by bacteriocin-producing bacteria, are a wonderful alternative to chemical preservatives in dairy products because they pose no health risks. Bacteriocins can be added to dairy products in purified/raw form, as a bacteriocin-producing LAB in the fermentation process, or as an adjuvant culture. Bacteriocins and bacteriocin-producing LAB have been used to control pathogens successfully in milk, yogurt, and cheeses in a number of cases. One of the most recent development is the inclusion of bacteriocins, whether directly as purified or semipurified form, or as bacteriocin-producing LAB, into bioactive films and coatings that are directly applied to food surfaces and packaging [53].

2.4.2 Probiotics

Probiotic is a relatively recent term that means "for life," and it refers to bacteria that have been connected to beneficial effects in humans and animals. The probiotic microorganisms are primarily *Lactobacillus* and *Bifidobacterium* strains, but *Bacillus, Pediococcus*, and several yeast strains have also been identified as suitable possibilities [54]. Sour/fermented milk, yogurt, cheese, butter/cream, ice cream, and infant formula all contain probiotic bacteria. These probiotics are either used as a starter culture alone or in combination with traditional starters, or incorporated into dairy products after fermentation, where their presence confers many functional characteristics to the product (such as improved aroma, taste, and textural characteristics), as well as many health-promoting properties [55].

Milk and milk products, particularly fermented dairy foods, are thought to be excellent carriers of probiotic strains, which allows to express their health-promoting functions to the greatest extent possible. Probiotic microorganisms can be concentrated and added in small amounts directly to food or a milk product, where they can grow. Yogurt is a well-known example of probiotic-rich functional dairy food. Probiotic yogurt, also known as bio-yogurt, should contain living bacterial cultures. Probiotics have been used to treat intestinal disorders as dietary supplements and oral agents. Probiotics have appeared recently as among the most precious bugs due to their ability to express a plethora of novel health-promoting functions that are strain-specific. Immunomodulation, restoring the balance of disturbed gut flora, strengthening the mucosal barrier function, and preventing lactose intolerance are the most notable probiotic functions. However, the focus at the moment is on researching probiotics as potential biotherapeutics for chronic inflammatory metabolic disorders such as diabetes, CVD, obesity, irritable bowel disease (IBD) and syndrome (IBS), Ulcerative Colitis (UC), Crohn's disease (CD), acute diarrhea, serum cholesterol reduction, shortening the duration of respiratory infections, blood pressure control, colon cancer, and urinary tract infection (UTI), among others.

2.4.3 Biotechnology and enzyme production

Enzyme production is a new field that answers the needs of the food processing industry by drastically lowering investment and processing costs. Enzymes are a biotechnological processing tool whose action in the food matrix may be manipulated to produce high-quality products. Moreover, the application of biotechnology has a significant role to produce enzymes used in the food and dairy industries, microbial protease, lipase, and galactosidase are enzymes that come from beneficial microorganisms. Their thermoresistance, thermostability, and thermoacidophilic qualities brought a particular interest to food producers [56].

The industrial production of enzymes for use in food processing dates back to 1874 when Danish scientist Christian Hansen extracted rennin (chymosin) from calves' stomachs for use in cheese manufacturing. Bovine chymosin was the first enzyme to be produced through biotechnological approaches in *E. coli*. Since then, genetic manipulation has been used to make tailor-made enzymes for specific consumer requirements. Now enzymes can be produced through recombinant DNA technology in large quantities for their subsequent application in the food industry (**Table 1**).

Some microorganism strains have been genetically modified to boost their capacity for enzyme synthesis under ideal conditions. In most situations, changed genes from other kingdoms of microorganisms can be found in GM microorganisms that generate enzymes. Bio-based compounds such as glucoamylase, lipase, –amylase, pectinase, antibiotics, amino acids, lactic acid, nucleic acid, and polysaccharides are created utilizing GM starting cultures. For example, one of the DNA codes for chymosin, which causes milk to curdle or coagulate during cheese fermentation, was cloned

Enzymes	Microorganisms	Application
Phytase	Aspergillus niger	Dair
Lipase		Baking, cheese flavor development
Lipase	Aspergillus oryzae	Cheddar cheese production
Chymosin	Escherichia coli K-12	Cheese
Chymosin	Kluyveromyces marxianus var. lactis	Cheese production
Microbial rennet	Mucor miehei	Baking, starch, cheese
Protease Starch,	Aspergillus usamii	brewing, meat tenderizer, milk coagulation, improvement of bread quality
β-Galactosidase (lactase)	LAB	Lactose intolerance reduction in people Prebiotic food ingredients

Source: Ghoshal [57].

Table 1.
Enzymes produced from genetically modified microorganisms using gene technology used in the dairy industry.

into bacteria (*Escherichia coli*), yeast (*Kluyveromyces lactis*), and mold (*Bacillus niger*) (*Aspergillus niger*). In Thailand, modified *E. coli* is being utilized to produce lysine, with the goal of increasing yield in less time [58].

2.5 Future application of biotechnology in the dairy industry

In the past, biotechnology has made substantial contributions to the dairy sector from animal genetic improvement to milk product processing. Thus, the following areas are some examples of possible applications of biotechnologies in future scenarios.

Dairy production

- Recombinant bovine
- Recombinant vaccines
- DNA fingerprinting
- Embryo transfer technology
- Animal cloning
- Gene forming and transgenic

Dairy processing

- Food grade bio-preservatives
- Dairy enzymes/proteins
- Probiotics

- Functional foods and nutraceuticals
- Dairy waste organization and pollution control

3. Conclusion

World dairy production and consumption were significantly increased in the last three decades, researchers and trend analysts claim that this growth will continue for the next few decades. However, in some countries especially in the EU, the number of cows is decreasing in recent years due to environmental, animal welfare, and other reasons. On the other hand, a number of technologies are invented and introduced to support the intensification of dairy farms. Recent technologies such as automatic milking machines, sensors, blockchain, and automatic feeders can provide significant improvements to milk production, environmental sustainability, and animal welfare in livestock agriculture. Similarly, sophisticated milk processing technologies have also been developed, which will have a remarkable role to produce dairy products that are wholesome and fit for human consumption. As well most recent processing technologies have a great potential for reducing GHG emissions during production, processing, and storage. Recently new biotechnological products are being developed for use in both animal production and food processing, dairy product bio-preservation, probiotics manipulation and synthesis, enzyme manufacturing, milk-derived bioactive peptides, other functional components production, and starter cultures technology and genetic manipulation are all examples of biotechnological applications. Conversely, the invention and application of these mysterious technologies were restricted in developed countries. Therefore, these advanced technologies should be more accessible to farmers around the world particularly to farmers in developing countries to improve milk production per cow, reduce higher GHG emissions, and feed growing populations.

Author details

Muhammed Nurye Gebeyehu
Department of Food Science and Postharvest Technology, Oda Bultum University College of Agriculture, Chiro, Ethiopia

*Address all correspondence to: muhammednurye86@gmail.com

References

[1] Nimbalkar V, Verma HK, Singh J. Dairy farming innovations for productivity enhancement. In: Qureshi MS, editor. New Advances in the Dairy Industry. London, United Kingdom: IntechOpen; 2021. DOI: 10.5772/intechopen.101373

[2] Datta N, Tomasula PM. Emerging Dairy Processing Technologies: Opportunities for the Dairy Industry. West Sussex, UK: John Wiley & Sons; 2015

[3] FAO. Gateway to dairy production and products. 2022. https://www.fao.org/dairy-production-products/production/en/

[4] Amiri S, Aghamirzaei M, Mostashari P, Sarbazi M, Tizchang S, Madahi H. The impact of biotechnology on dairy industry. In: Applied Biotechnology Reviews, Microbial Biotechnology in Food and Health. London, UK: Academic Press; 2021. pp. 53-79. DOI: 10.1016/B978-0-12-819813-1.00003-7

[5] Gomes AA, Braga SP, Cruz AG, Cadena RS, Lollo PC, Carvalho C, et al. Effect of the inoculation level of Lactobacillus acidophilus in probiotic cheese on the physicochemical features and sensory performance compared with commercial cheeses. Journal of Dairy Science. 2011;**94**:e4777-e4786

[6] Minervini F, Siragusa S, Faccia M, Dal Bello F, Gobbetti M, De Angelis M. Manufacture of Fior di Latte cheese by incorporation of probiotic lactobacilli. Journal of Dairy Science. 2012;**95**:e508-e520

[7] OECD-FAO. Agricultural Outlook 2020-2029. 2020

[8] AHDB (Agriculture and Horticulture Development Board). 2022. Dairy market outlook

[9] Gindt K. Dairy Situation and Outlook. USA: The Board of Regents of the University of Wisconsin System; 2022

[10] European Commission. EU Agricultural Outlook for Markets, Income, and Environment, 2021-2031. Brussels: DG Agriculture, and Rural Development; 2021

[11] FAO. Global dairy industry and trends. International Magazine for Animal Feed & Additives Industry. 2021

[12] Minj J, Aparna SV, Kumari A. Dairy Processing: Advanced Research to Applications. Singapore: Springer; 2020

[13] Wondatir. Livestock production systems in relation with feed availability in the highlands and central rift valley of Ethiopia. 2010

[14] Akbar MO, Shahbaz Khan MS, Ali MJ, Hussain A, Qaiser G, Pasha M, et al. IoT for development of smart dairy farming. Journal of Food Quality. 2020;**2020**:4242805. DOI: 10.1155/2020/4242805

[15] Mishurov NP. Information management of dairy cattle breeding. Bulletin of VNIIMZh. 2014;**4**(16):41-48

[16] Tsoi YA, Mishurov NP. State and development trends of robotic equipment for milking cows. Rural Machinery and Equipment. 2019;**5**(263):2-9

[17] Viguier C, Arora S, Gilmartin N, Welbeck K, O'Kennedy R. Mastitis detection: Current trends and future

perspectives. Trends in Biotechnology. 2009;**27**(8):37

[18] Lessire F, Moula N, Hornick J, Dufrasne I. Systematic review and meta-analysis: Identification of factors influencing milking frequency of cows in automatic milking systems combined with grazing. Animals. 2020;**10**:913

[19] Masía FM, Lyons NA, Piccardi M, Balzarini M, Hovey RC, Garcia SC. Modeling variability of the lactation curves of cows in automated milking systems. Journal of Dairy Science. 2020;**103**:8189-8196

[20] Edwards JP, Kuhn-Sherlock B, Dela Rue BT, Eastwood CR. Short communication: Technologies and milking practices that reduce hours of work and increase flexibility through milking efficiency in pasture-based dairy farm systems. Journal of Dairy Science. 2020;**103**:7172-7179

[21] Karttunen JP. Occupational health and safety of Finnish dairy farmers using automatic milking systems. Frontiers in Public Health. 2016;**4**:1-11. DOI: 10.3389/fpubh.2016.00147

[22] Salfer JA, Siewert JM, Endres MI. Housing, management characteristics, and factors associated with lameness, hock lesion, and hygiene of lactating dairy cattle on Upper Midwest United States dairy farms using automatic milking systems. Journal of Dairy Science. 2018;**101**:8586-8594. DOI: 10.3168/jds.2017-13925

[23] Hansen BG, Stræte EP. Dairy farmers' job satisfaction and the influence of automatic milking systems. Journal of Life Science. 2020;**92**:100328

[24] Friggens NC, Blanc F, Berry DP, Puillet L. Review: Deciphering animal robustness. A synthesis to facilitate its use in livestock breeding and management. Animal. 2017;**11**:2237-2251

[25] Göncü S, Güngör C. The innovative techniques in animal husbandry. In: Yucel B, editor. Animal Husbandry and Nutrition. London, United Kingdom: IntechOpen; 2018. DOI: 10.5772/intechopen.72501

[26] Grothmann A, Nydegger F, Moritz C, Bisaglia C. Automatic feeding systems for dairy cattle—Potential for optimization in dairy farming. In: International Conference on Agricultural Engineering: Towards Environmental Technologies. 2010

[27] Tikhomirov IA, Skorkin. Experience of using milking robots in dairy cattle breeding on the example of farms in the Kaluga region. Bulletin of VNIIMZh. 2019;**1**(33):160-165

[28] Marcos L, Filho S, Lopes MA, Brito SC, Rossi G, Conti L, et al. Robotic milking of dairy cows: A review (Ordenha robotizada de vacas leiteiras: Uma revisão). Semina Ciências Agrárias. 2020;**41**:2833-2850

[29] Hansen BG, Herje HO, Höva J. Profitability on dairy farms with automatic milking systems compared to farms with conventional milking systems. International Food and Agribusiness Management Review. 2019;**22**:215-228

[30] Goff HD, Griffiths MW. Major advances in fresh milk and milk products: Fluid Milk products and frozen desserts. Journal of Dairy Science. 2006;**89**(4):1163-1173. DOI: 10.3168/jds.S0022-0302(06)72185-3

[31] Shrivastava SL, Banerjee S. Recent trends in milk processing—A short review. Approaches in Poultry, Dairy & Veterinary Sciences. 2017;**2**(1):11-13. DOI: 10.31031/APDV.2017.02.000527

[32] Dolatowski ZJ, Stadnik J, Stasiak D. Applications of ultrasound In food technology. Acta Scientiarum Polonorum Technologia Alimentaria. 2007;**6**(3):89-99

[33] Reena, Kumar A. Thermal and non-thermal treatment of milk: A review. International Research Journal of Engineering and Technology (IRJET). 2021;**8**(5):2330-2332

[34] Singh AK, Borad S, Meena GS, Sharma H, Arora S. High-pressure processing of milk and milk products. In: Chauhan OP, editor. Non-thermal Processing of Foods. 1st ed. Boca Raton, Florida, United States: CRC Press, Taylor & Francis Group; 2019. pp. 69-88. Available from: https://lccn.loc.gov/2018033439

[35] Sampedro F, Rodrigo D. Pulsed electric fields (PEF) processing of milk and dairy products. In: Datta N, Tomasula PM, editors. Emerging Dairy Processing Technologies: Opportunities for the Dairy Industry. West Sussex, UK: John Wiley & Sons; 2015. pp. 115-147

[36] Zhang Q, Barbosa-Canovas GV, Swanson BG. Engineering aspects of pulsed electric field pasteurization. Journal of Food Engineering. 1995;**25**:261-281

[37] Jaeger H, Janositz A, Knorr D. La Réaction de Maillard et Son Contrôle Pendant La Fabrication Des Aliments. Le Potentiel Des Nouvelles Technologies. Pathologie et Biologie. 2010;**58**:207-213

[38] Mosqueda-Melgar J, Elez-Martínez P, Raybaudi-Massilia RM, Martín-Belloso O. Effects of pulsed electric fields on pathogenic microorganisms of major concern in fluid foods: A review. Critical Reviews in Food Science and Nutrition. 2008;**48**:747-759

[39] Joannes C, Sipaut CS, Dayou J, Yasir SM, Mansa RF. The potential of using pulsed electric field (PEF) technology as the cell disruption method to extract lipid from microalgae for biodiesel production. International Journal of Renewable Energy Research. 2015;**5**:598-621

[40] Kandušer M, Belič A, Čorović S, Škrjanc I. Modular serial flow through device for pulsed electric field treatment of the liquid samples. Scientific Reports. 2017;**7**:8115

[41] Schottroff F, Knappert J, Eppmann P, Krottenthaler A, Horneber T, McHardy C, et al. Development of a continuous pulsed electric field (PEF) Vortex-flow chamber for improved treatment homogeneity based on hydrodynamic optimization. Frontiers in Bioengineering and Biotechnology. 2020;**8**:340

[42] Ivorra A, Villemejane J, Mir LM. Electrical modeling of the influence of medium conductivity on electroporation. Physical Chemistry Chemical Physics. 2010;**12**:10055-10064

[43] Chandrapala J. Ultrasound processing of milk and dairy products. In: Ashokkumar M, editor. Handbook of Ultrasonics and Sonochemistry. Singapore: Springer; 2015. DOI: 10.1007/978-981-287-470-2_66-1

[44] Chávez-MartínezA,Reyes-VillagranaRA, Rentería-MonterrubioAL,Sánchez-VegaR, Tirado-Gallegos JM, Bolivar-Jacobo NA. Low and high-intensity ultrasound in dairy products: Applications and effects on physicochemical and microbiological quality. Food. 2020;**9**(11):1688. DOI: 10.3390/foods9111688

[45] Fernández A, Thompson A. The inactivation of Salmonella by cold atmospheric plasma treatment. Food Research International. 2012;**45**: 678-684

[46] Misra NN, Koubaa M, Roohinejad S, Juliano P, Alpas H, Inácio RS, et al. Landmarks in the historical development of twenty-first century food processing technologies. Food Research International. 2017;**97**:318-339

[47] Misra NN, Tiwari BK, Raghavarao KSMS, Cullen PJ. Nonthermal plasma inactivation of food-borne pathogens. Food Engineering Reviews. 2011;**3**:159-170

[48] Rathod NB, Kahar SP, Ranveer RC, Annapure US. Cold plasma an emerging non-thermal technology for milk and milk products: A review. International Journal of dairy technology. 2021;**74**(4):615-626

[49] Henning DR, Baer RJ, Hassan AN, Dave R. Major advances in concentrated and dry milk products, cheese, and milk fat-based concepts. Journal of Dairy Science. 2006;**89**:1179-1188

[50] Kumar P, Sharma N, Ranjan R, Kumar S, Bhat ZF, Jeong DK. Perspective of membrane technology in dairy industry: A review. Journal of Animal Science. 2013;**26**(9):1347-1358

[51] Sundarraj AA, Rajathi AA, Vishaal SC, Rohit D, Prakash MS, Sam AA, et al. Food biotechnology applications in dairy and dairy products. Journal of Pharmacy Research. 2018;**12**(4):520-525

[52] Padmavathi. "Modern biotechnology"—A tremendous application over today's dairy industry. International Journal of Advanced Studies. 2018;**3**(3):9

[53] Silva CCG, Silva SPM, Ribeiro SC. Application of bacteriocins and protective cultures in dairy food preservation. Frontiers in Microbiology. 2018;**9**(9):594

[54] Soccol CR, de Souza VL, Rigon SM, Pedroni MA, Yamaguishi CT, de Deae LJ, et al. The potential of probiotics: A review. Food Technology and Biotechnology. 2010;**48**:413-434

[55] Gao J, Li X, Zhang G, Sadiq FA, Simal-Gandara J, Xiao J, et al. Probiotics in the dairy industry—Advances and opportunities. Comprehensive Reviews in Food Science and Food Safety. 2021;**20**(4):3937-3982. DOI: 10.1111/1541-4337.12755

[56] Al-Manhel AJA. Application of microbial enzymes in dairy products: A review. Basrah Journal of Agricultural Science. 2018;**31**(1):20-30

[57] Ghoshal G. Biotechnology in food processing and preservation: An overview. In: Wolff AG, editor. Advances in Biotechnology for Food Industry. Handbook of Food Bioengineering. London, United Kingdom: Academic Press, Elsevier; 2018. pp. 27-54

[58] Enitan-Folami AM, Swalaha FM. 10 Application of biotechnology in the food industry. In: Ijabadeniyi OA, editor. Food Science and Technology: Trends and Future Prospects. Berlin, Boston: De Gruyter; 2020. pp. 235-277. DOI: 10.1515/9783110667462-010

Edited by Shumaila Manzoor and Muhammad Abubakar

Livestock farming supports livelihood and provides food security. It is the fastest-growing sector of the agriculture economy. This book is about using modern technology to increase yields, income, and ultimately food security. It is organized into three sections on livestock and poultry farming, fish farming, and innovations and advances in technology.

Published in London, UK

IntechOpen

ISBN 978-1-80356-103-5