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Reduced Meat Consumption: from Multicriteria Argument Modelling to Agent-Based Social Simulation

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Abstract

A second nutrition transition seems to be emerging towards more plant-based diets, curbing meat consumption in developed countries at the beginning of the 21st century. This shift suggests that rational arguments tend to influence an increasing number of individuals to adopt vegetarian diets. This work aimed to understand and simulate the impact of different types of messages on the choice to change food diets at the individual level, and the impact of the diffusion of opinions at the collective level. It provided two results: (1) a network of arguments around vegetarian diets is modelled using an abstract argumentation approach. Each argument, formalized by a node, was connected with other arguments by arrows, thus formalizing relationships between arguments. This methodology made it possible to formalize an argument network about vegetarian diets and to identify the importance of health arguments compared to ethical or other types of arguments. This methodology also identified key arguments as a result of their high centrality in being challenged or challenging other arguments. The results of constructing this argument network suggested that any controversy surrounding vegetarian diets will be polarized around such high centrality arguments about health. Even though few ethical arguments appeared in our network, the health arguments concerning the necessity or not of animal products for humans were indirectly connected with ethical choices towards vegetarian diets; (2) an agent-based simulation of the social diffusion of opinions and practices concerning meat consumption is then introduced. The purpose of this simulation was to capture the balance of vegetarian vs. meat-based diets. It contributes to modelling consumer choices by exploring the balance between individual values and external influences such as social pressure, communication campaigns and sanitary, environmental or ethical crises.

Keywords: Sustainable food systems; Agent-based modelling; Argumentation systems; Food diet; Consumer choice criteria; Animal food product

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1 Introduction

The first nutrition transition involved the increase of sugar, fat, meat and processed products in human diets (Popkin, 1993) and is the dominant nutritional model today. Vranken, Avermaete, Petalios and Mathijs (2014) identified a second nutrition transition that is occurring in the most developed countries where meat consumption is currently decreasing. The transition towards the reduction of meat consumption covers a wide variety of practices ranging from occasional vegetarianism to veganism (also called "strict vegetarian") (Beardsworth & Keil, 1991). The rationale for such a transition mainly implies ethical and health concerns (Jabs, Devine & Sobal, 1998), but the environmental impact of meat consumption is also highlighted to a lesser degree (Ruby, 2012). McDonald (2000) conducted individual interviews with vegans and found that their nutrition transition depended on a catalytic experience that oriented individuals towards information acquisition and ultimately led to a decision to change diets. However, the information that led to the decision is not precisely known among vegetarians. Under what conditions are we likely to observe the emergence of a second transition? This paper aims to understand and simulate the impact of different types of messages on the choice to change food diets at the individual level, and the impact of the diffusion of opinions at the collective level.

As a first step, we present a methodology to explore the main arguments and the relationships between them that transitioning individuals may face. The approach is based on the principles of argumentation networks, an artificial intelligence model based on the construction and evaluation of interacting arguments. Most of the existing models are grounded in the abstract argumentation framework proposed in Dung (1995). In this framework, an argumentation system is defined by a set of arguments A, and an attack relation between arguments R. Sets of arguments that "make sense" together are then computed and are referred to as extensions. In a subsequent study (Thomopoulos, 2018), various indicators were defined to analyze argumentation systems by highlighting consensual and non-consensual aspects of the issue debated.

As a second step, an agent-based simulation is proposed. Following the theoretical approach of Xie et al. (2011), argument networks may take advantage of being used together with agent-based modelling (ABM) to explore the emergent establishment of new social norms in the specific case of vegetarianism. Such a model can help understand the conditions under which such arguments could circulate in a population and favour the adoption of a vegetarian diet.

Agent-based models offer a way of taking inter-individual heterogeneity, social interaction and interdependence, adaptation, and decision-making into account at different levels. In the environmental sciences, these models have proved to be a way to capture complex characteristics of socio-ecological systems (An, 2012; Berger, 2001; Epstein & Axtell, 1996; Filatova, Verburg, Parker & Stannard, 2013; Matthews, Gilbert, Roach, Polhill & Gotts, 2007; Parker, Manson, Janssen, Hoffmann & Deadman, 2003; Rammel, Stagl & Wilfing, 2007). In this family of ABM studies, ecology and geography are dominant (Castella, Trung & Boissau, 2005; Grimm, 1999; Parker et al., 2003; Railsback, Lytinen & Jackson, 2006; Verburg & Veldkamp, 2005). However, the social sciences are also present in ABM approaches to assess and explain the complexity of human decision-making processes and behaviours (Epstein & Axtell, 1996; Janssen, Walker, Langridge & Abel, 2000; Janssen & Ostrom, 2006; Robinson et al., 2007; Schelling, 1971; Simon, 1997).

In the food sciences, historically based on process-oriented studies, ABMs are absent from the range of models used to approach food quality and, as a more recent concern, food sustainability. Recent studies related to multi-agent systems applied to the agri-food sector are those based on the argumentation theory (Bourguet, Thomopoulos, Mugnier & Abecassis, 2013; Thomopoulos, Croitoru & Tamani, 2015; Thomopoulos, Moulin & Bedoussac, 2017). However, social issues are deeply involved in food system sustainability. Consumer demand, environmental awareness, willingness to pay, acceptability of products and the transmission of new food habits are all key factors to analyze the food system. Consequently, there is an urgent need and a real challenge to take food-related social

behaviors into account and to integrate them into food policy analysis.
A previous approach for combining argumentation and simulation for decision support in food systems was proposed in Thomopoulos et al. (2017). The simulation model considered in that study was systems dynamics. In this paper, an ABM simulation of the social diffusion of opinions and practices concerning meat consumption is proposed. It aims to capture the balance of vegetarian vs. meat food diets. More specifically, we aim to understand the balance between individual values and external influences such as social pressure, communication campaigns and sanitary, environmental or ethical crises in the decision-making of individuals with regard to their food diet.

2 Materials and Methods

2.1 Argumentation Formalism

In order to model arguments involved in vegetarian transitions, we used an abstract argumentation approach (Dung, 1995; Rahwan & Simari, 2009; Thomopoulos, 2018). An argumentation system is usually represented as an oriented graph where nodes are arguments and edges are attack relations between arguments (Figure 1). Considering Dung (1995) seminal work on argumentation, an argument and the attack relation are abstract and can be instantiated and defined in different ways in different contexts (Walton & Macagno, 2015). Dung himself stated: "an argument is an abstract entity whose role is solely determined by its relations to other arguments. No special attention is paid to the internal structure of the arguments." For example, an argument can be a set of statements composed of a conclusion and at least one premise, linked by an inference or a logical relationship. Attacking an argument can be achieved in different ways:

1. by raising doubts about its acceptability through critical questions;
2. by questioning its premises;
3. by suggesting that the premises are not relevant to the conclusion; or
4. by presenting an argument with an opposing conclusion.

In all these cases, an attack relation is said to exist.
Even though Dung's framework is theoretically sound, it is not straightforward enough to be applied to real-life situations. Indeed, one of the initial difficulties is how to define an argument in order to properly reflect stakeholders' statements in a debate. Unfortunately, there is still no general model that can be used to formalize a natural argument (i.e., an argument stated by a stakeholder during a discussion in natural language) and input in an abstract argumentation framework in a real decision-making context. Quoting Baroni and Giacomin (2009): "While the word 'argument' may recall several intuitive meanings, like the ones of 'line of reasoning leading from some premise to a conclusion' or of 'utterance in a dispute', abstract argument systems are not (even implicitly or indirectly) bound to any of them: an abstract argument is not assumed to have any specific structure but, roughly speaking, an argument is anything that may attack or be attacked by another argument." Indeed, the structure of an abstract argument does not correspond to the intuitive understanding of what an argument is. Moreover, the notion of "attack between arguments" does not have a natural and direct correspondence to practical expressions used by stakeholders when debating. Finally, representing arguments as an oriented graph can be a difficult task for stakeholders: when the number of arguments and/or attacks is large, the graph becomes illegible and difficult to interpret by the stakeholders. For all these reasons, the ways that arguments and attacks are identified and modelled is part of the project contributions. They are thus presented in Section 3.

The arguments

In our project, we needed to find a practical way of defining arguments that are used in the process of decision-making. In such a context, arguments can be intuitively thought of as being statements to support, contradict or explain opinions or decisions (Amgoud & Prade, 2009). More precisely,

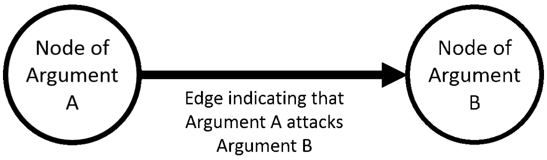

Figure 1: General graphical representation of an argumentation system

in decisional argumentation frameworks (Ouerdane, Maudet & Tsoukiàs, 2010), the argument definition is enriched with additional features, namely the decision (also referred to as 'action', 'option' or 'alternative') and the goal (also referred to as 'target'). In other studies, arguments are also associated with specific actors. An application of a decision-oriented argumentation framework to a real-life problem concerning food policy can be found in Bourguet et al. (2013), where a recommendation regarding the provision of whole-grain bread was analyzed *a posteriori*. In this case, each argument is associated with the action it supports.

The attack relation

Now, let us consider the attack relation. In structured argumentation (i.e., logic-based argumentation frameworks where arguments are obtained as instantiations over an inconsistent knowledge base) three kinds of attacks have been defined: undercut, rebut and undermine (Besnard & Hunter, 2008). The intuition of these attack relations is either to counter the premise of the opposing argument ('undercut'), the conclusion ('rebut') or to attack the logical steps that allowed the inference between the argument's premise and conclusion ('undermine'). In abstract argumentation, the set of attacks is simply considered as provided a priori. Another possibility that can be considered is to enhance the argumentation framework with a set of preferences expressed, for instance, as weights representing uncertainty. In our project, we needed to choose a practical way to define the attack relation.

2.2 Agent-Based Modeling

Agent-based models are computational models used to simulate the actions and interactions of individual or collective autonomous agents in order to assess their effects on the system as a whole. They attempt to reproduce and predict the emergence of complex phenomena induced from the micro-scale to the macro-scale. Their principle is that simple local behavioural rules generate complex global behaviour. An overview of their early history can be found in Samuelson (2000), and more recent developments in Samuelson and Macal (2006). The model introduced was built and run on two different agent-based modelling and simulation platforms, namely CORMAS and GAMA (Taillandier et al., 2018). The CORMAS platform (http://cormas.cirad.fr/) was specifically developed by CIRAD – the French international cooperation organization for agricultural research in the global South – to simulate natural resource management, and is oriented towards the representation of interactions between stakeholders about resource use. In CORMAS, entities are categorized into three types: spatial entities describing the space at different aggregation levels, passive entities that are objects that can be manipulated by social agents, and social entities that can make decisions, move, and interact with other agents. The GAMA platform platform (http://gama-platform.org) was developed by a consortium of research teams using the JAVA language. It is generalist in its application domains and is particularly well-suited for connection with geographical information system data and visualization.

3 Results and Discussion

3.1 Argumentation Results

Contributions to Modelling Arguments and Attacks

We introduced the specification of an argument as a tuple a = (I;T;S;R;C;A;Is;Ts) where:

I was the identifier of the argument;

T was the type of the argument (with values in favor of, denoted by '+', or against, denoted by '-', the vegetarian option);

S was the statement of the argument, i.e., its conclusion;

R was the rationale underlying the argument, i.e., its hypothesis;

C was the criterion on which the argument relies;

A was the actor who proposes the argument;

Is was the information source containing the argument;

Ts was the type of source the argument comes from.

For any argument a, we denoted by I(a), T(a), S(a), R(a), C(a), A(a), Is(a), Ts(a), the identifier, the type, the statement, the rationale, the criterion, the actor, the information source and the information type of argument a, respectively.
Considering the reality of stakeholders' debates and our model to formalize arguments, we chose to model the attack relation in the following way. Attacking an argument a was achieved by:

1 explicitly raising doubts about its acceptability by expressing a counter-argument citing a or the information source containing a;

2 implicitly raising doubts about its acceptability by expressing a counter-argument contradicting a through undercut, rebut or undermine. Formally, we considered the following attack relation:

Let a and b be two arguments. Then, a attacks b if and only if the following two conditions are satisfied:

$$T(a) \neq T(b) \quad (1)$$
$$\{R(a), S(a)\} \Rightarrow not\{R(b), S(b)\} \quad (2)$$

The first condition expresses the fact that one of the arguments a and b is in favour and the other against the vegetarian option. The second condition expresses the inconsistency of a and b, i.e., if the hypothesis and the conclusion of argument a are assumed to be true, then the hypothesis of b or the conclusion of b, or both, do not stand.

Exploring the Literature

Following the above formalism, we extracted arguments in favour or not of reducing animal product consumption. Our sources of arguments were newspapers, grey literature and the top ten Google search words ("vegetarian diet"; "vegan diet"; "vegetalism argument"). The latter inquiry was added to the pool of popular scientific papers, WebMedia articles and blog posts. We thoroughly read each source and extracted all of the arguments as expressed by their authors. For each argument, we attributed a criterion ("Nutritional"; "Economic"; "Environmental"; "Anthropological"; "Ethical"; "Health" or "Social") and noted the source expressing this argument ("Journalist"; "Scientist", "Philosopher"; "Blogger", etc.). We consequently obtained 114 arguments.
Table 1 displays a sample of the set of arguments considered in our case study. Each argument was first formalized by an identification number, whether it was in favour or not of meat reduction diets (+/-), its main statement and rationale such as: "Vegan diet is related to vitamin B12 deficiency" (Statement) since "plants do not contain vitamin B12" (Rationale). Other information (Actor, Information source & Type of source) characterized the origin of the argument. Based on this first step, we then formalized attacks between them. An attack occured when an argument contradicted another one. For example, the argument "1" quoted above is contradicted by the following argument "28 - Properly planned vegetarian or vegan diets fit all stages of life" since "Nutrient needs are satisfied and growth is normal." When these arguments are graphically formalized, each one is represented as a node, and an attack is a vertex connecting both arguments where the arrow points to the direction of the attack. In our case study, we identified 155 attacks connecting 55 arguments out of a total of 114.

Table 1: Sample of Argument Tuples about Reduced Meat Consumption (I: Identification; T: Type; Is: Information source; Ts: Type of source)

I	T	Statement	Rationale	Criterion	Actor	Is	Ts
1	-	Vegan diets are deficient in vitamin B12	Plant proteins do not contain vitamin B12	Nutritional	Journalist	Canard Enchainé - 144 - July 2017	Newspaper
7	+	Persons not consuming animal products do not have health issues related to proteins	Combined protein intakes ensure a balanced diet	Nutritional	Innovation cluster	Valorial	PowerPoint
15	-	Plant proteins trigger allergies	Plant-based food are more often allergenic	Nutritional	Innovation cluster	Valorial	PowerPoint
23	+	Vegetarian diets are good for health	Diabetes, cancer and coronary risks are reduced	Health	Scientists	Tilman and Clark (2014)	Scientific paper
28	+	Properly planned vegan or vegetarian diets fit all stages of the life cycle	Nutrient needs are satisfied and growth is normal	Health	American Dietetic Association	Craig and Mangels (2009)	Scientific paper
43	+	Vegan diets improve rheumatoid arthritis activity	A diet-induced fecal flora change was observed	Health	Scientists	Peltonen et al. (1997)	Scientific paper
55	+	Eating meat is not in human nature	It was sometimes necessary in the past to eat meat, not today	Anthropological	Blogger pro-vegan	Blog - Eleusis and Megara	Blog post
56	+	Stopping eating animals does not mean animal extinction	Deforestation for the cultivation of animal feed provokes species extinctions	Environmental	Blogger pro-vegan	Blog - Eleusis and Megara	Blog post
57	+	Animal products are not essential to good health	B12 intakes in animal products come from B12 industry production anyway	Nutritional	Blogger pro-vegan	Blog - Eleusis and Megara	Blog post
71	-	No study is favourable to the vegan diet	One good-quality study showed that the Atkins diet is better than the Ornish diet	Health	Journalist	Signs of the Times	Internet article
72	-	No study is favourable to the vegan diet	Study showing health and mortality advantages from a vegetarian and vegan diet do not show causation	Health	Journalist	Signs of the Times	Internet article
77	-	No health reason justifies avoiding animal products	The human body has been adapted to eating animal products for millions of years	Health	Journalist	Signs of the Times	Internet article
83	-	Vegan diet safety is not proven	So far, scientific studies do not have much value and especially on long-term effects.	Health	Philosopher	Huffington Post	Blog post
108	+	Animal products are useless for your health	The American Dietetic Association says so	Nutritional	Website editor	VeganFrance.fr	Web directory
111	+	There is no health issue with a balanced vegan diet	It is recognized by many governmental and non-governmental organizations. Dieticians from Canada even say it has advantages for health	Health	Philosopher	Blog – Frédéric Côté-Boudreau	Blog post

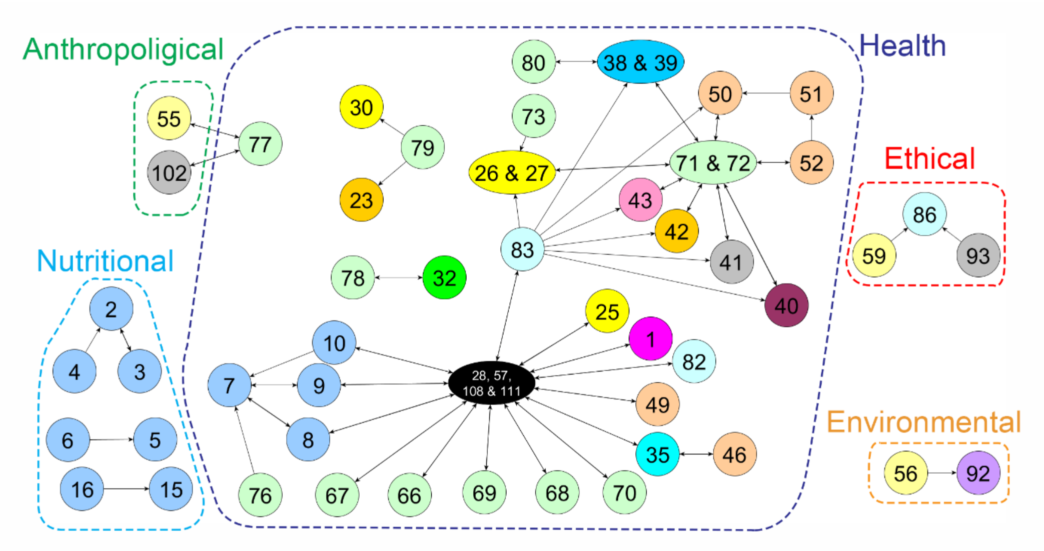

Figure 2: Graphical Representation of Arguments and Attacks about Reduced Meat Consumption

Graphical representation of the argument network

In order to make a graphical representation of the argument network, we used the visualization program Yed Graph Editor (version 3.17.1). We chose to represent only arguments that were connected with at least one attack. Hence, 55 arguments were kept and 59 were rejected. The result is displayed in Figure 2. Each number corresponds to one argument expressed by one source. Arguments were grouped in dashed lines according to the criterion they were based on, which is indicated in the legends (Nutritional, Ethical, etc.). Each argument node received a specific colour according to its source (e.g., pink for argument 1 from the newspaper "Le Canard Enchainé", No. 144, published in July 2017, blue for arguments 2 to 16 from the Valorial competitiveness cluster's presentation, etc.). For visualization purposes, we merged identical arguments in the same node when they were repeated and came from the same source.

3.2 Agent-Based Simulation Results

Conceptual model

Scenario Description

The system modelled was a country consisting of a population of N citizens. Each citizen was characterized by: (i) a constant level of need for quality regarding food (environmental preservation, ethics, health, taste, etc.); (ii) a variable level of perception of meat products corresponding to these criteria. Depending on these two levels, citizens had a behaviour regarding their food diet - either meat consumers if their perception exceeded their needs, or vegetarian if their needed exceed their perception. This principle is represented in Figure 3. Moreover, each citizen had some resistance to change, represented by the resistance rate.
Within this system, each citizen communicated with his/her direct neighbours and may then have changed his/her perception of meat consumption and, consequently, his/her diet. Citizens were also influenced by advertising campaigns that target every citizen and tend to increase meat product perceptions in the whole

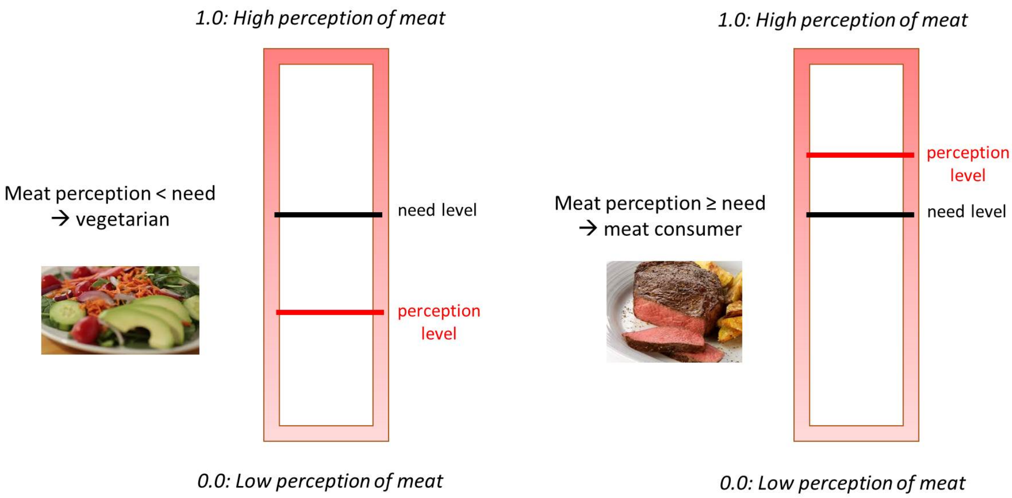

Figure 3: Behavioural model of food diet choice

country. Finally, crises broke out with a given frequency. These represented, in particular, sanitary crises and, more generally, ethical or environmental crises. All of the citizens received the information, all the more since they were spatially close to the crisis event. Their perception of meat products may have been impacted.
Initial parameters subject to stochasticity included the distribution of need and perception levels in the population with respect to some initial proportions of vegetarians and meat consumers. The output parameter observed was the evolution of food diets over time.

Working Hypotheses

The ABM developed was based on the following assumptions:

- A cognitive theory of food perceptions and needs. This was where the ABM was related to arguments in favour or not of a diet, and where the importance attached to the criteria underlying arguments for a given diet was expressed as a level of needs. The failure of a food diet to be perceived as being consistent with these needs led to a behavioural change in the ABM, which could not be captured by the argumentation system alone.
In Schluter et al. (2017), six categories of human decision-making behaviours in socio-ecological systems are described. In the present paper, we drew on the "habitual behavior", described by the following characteristics (quoting Schluter et al. (2017)):

'Behavior is initially deliberate and goal-directed'
'if new behaviour is rewarded, the chances increase that it will be repeated'
'repeatedly obtaining satisfactory rewards reinforces the behaviour'
'the selection of behaviour will be automatic as long as needs are satisfied'
'the actor will stop automatic behaviour and deliberate about alternative behaviours if need satisfaction drops below a critical level'
'if the reward devalues or disappears, habitual behaviour persists at first, but will go extinct after longer absence of reward.'

- Three triggers for vegetarian/meat product demand:

  – occurrence of sanitary, environmental

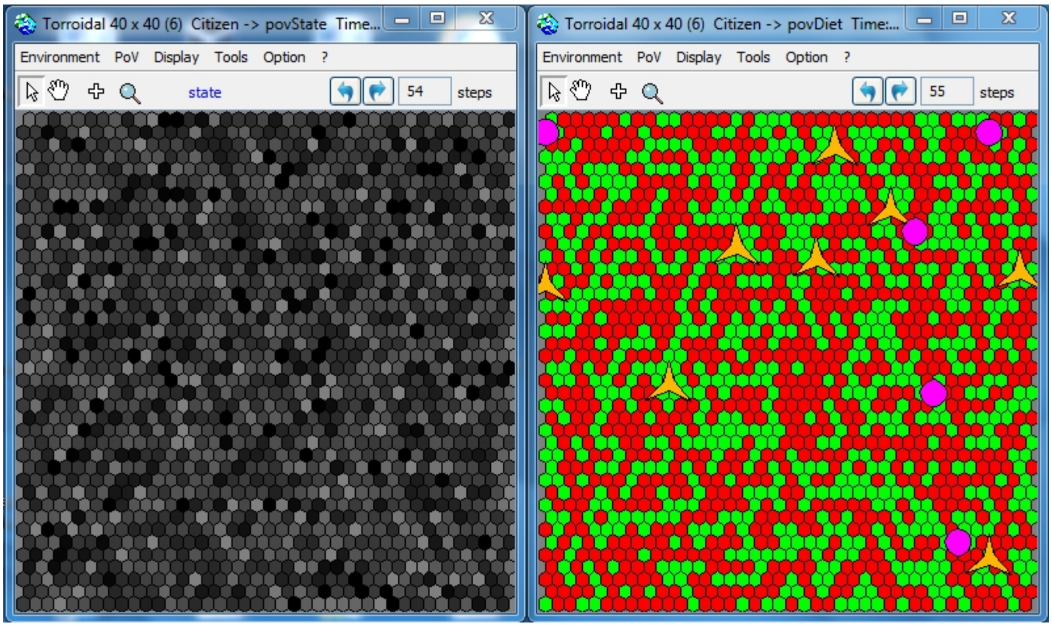

Figure 4: A running simulation in CORMAS

and ethical crises;

- neighbourhood effect;
- advertising campaigns.

The influences of these triggers were assumed to follow the opinion diffusion model (Deffuant, Amblard, Weisbuch & Faure, 2002). An opinion (in this case, a perception of meat products) was represented by a numerical float value between 0.0 and 1.0, initialized with a random value that was less than needs for vegetarians, and greater than needs for meat consumers. At each step, agents consulted their neighbours and updated their opinions as follows. When agent x consulted his/her n neighbours yi (i ϵ [1; n]), his/her new opinion *op* became:

$$op_x(t+1) = rR * op_x(t) + (1 - rR) * \overline{op_{y_i}(t)}_i \tag{3}$$

The resistance rate rR was a parameter of the simulation, i.e., a number in [0.0, 1.0] and $\overline{op}$ is the mean opinion.

Simulations Results

Figure 4 displays a running simulation of the model developed, implemented in the CORMAS platform. The left part shows consumer perceptions of meat products (the darker, the higher). The right side displays the resulting food diets: green for vegetarians, red for meat consumers. The yellow stars depict the occurrence of a crisis that tends to decrease people's perceptions of meat products, all the more since the crisis localization is close. The pink circles depict the occurrence of an advertising campaign that tends to increase people's perceptions of meat products. Although localized (in order to visualize them), advertising campaigns were nationwide and impacted every citizen, as already mentioned.
Figure 5 shows a running simulation of the same model, implemented in the GAMA platform. As in CORMAS, food diets are represented in green for vegetarians and in red for meat consumers. Perceptions of meat products are represented by the sizes of the circles (the larger the circle, the higher the perception). The left part of the screen makes it possible to display and easily modify simulation parameters.

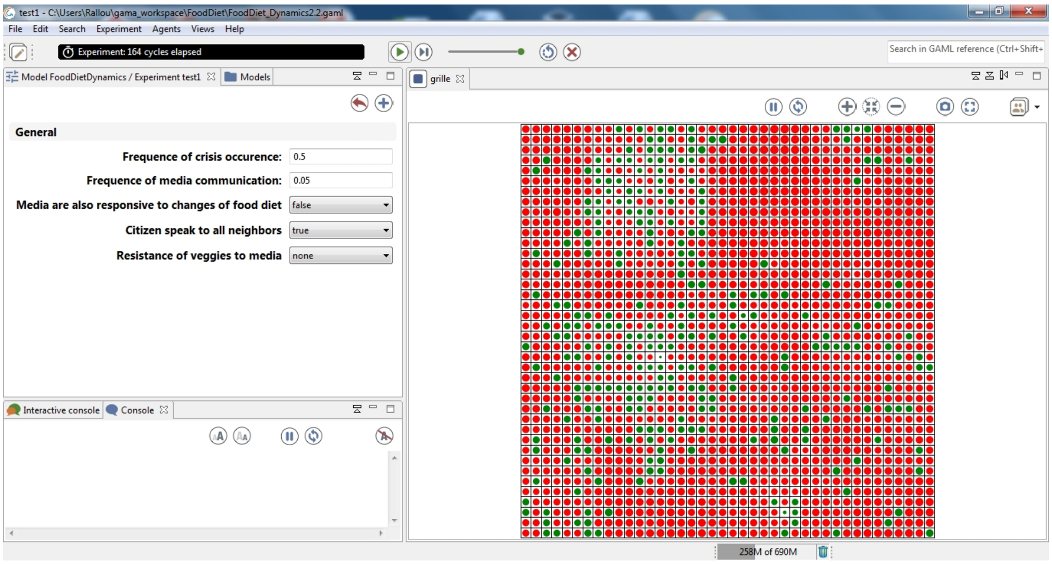

Figure 5: A running simulation in GAMA

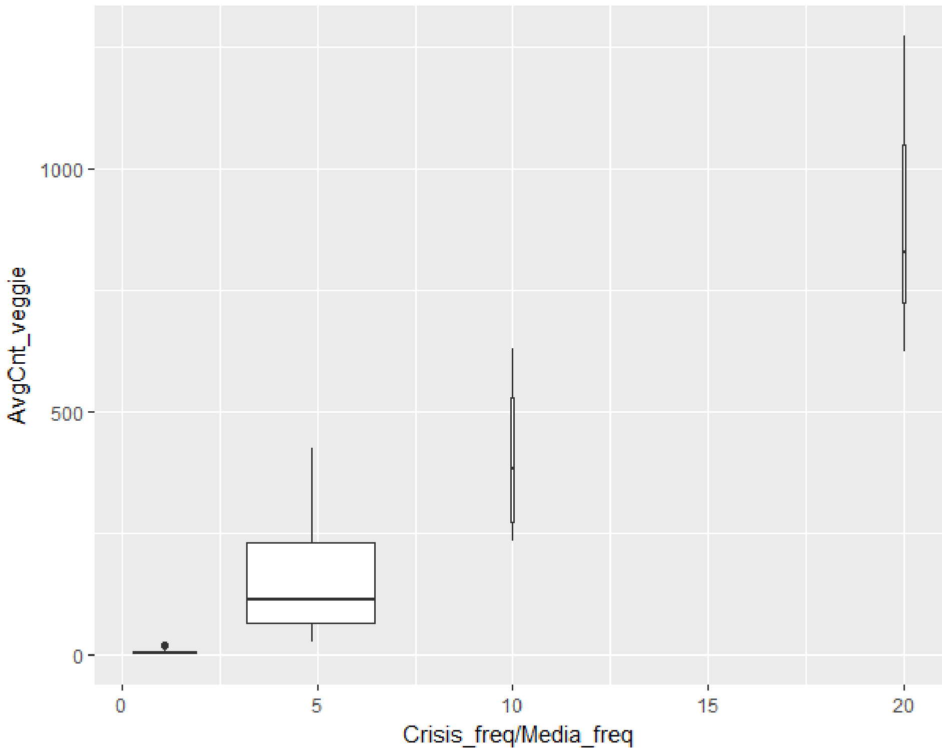

Figure 6: Effect of the ratio between the frequency of crises and that of communication campaigns

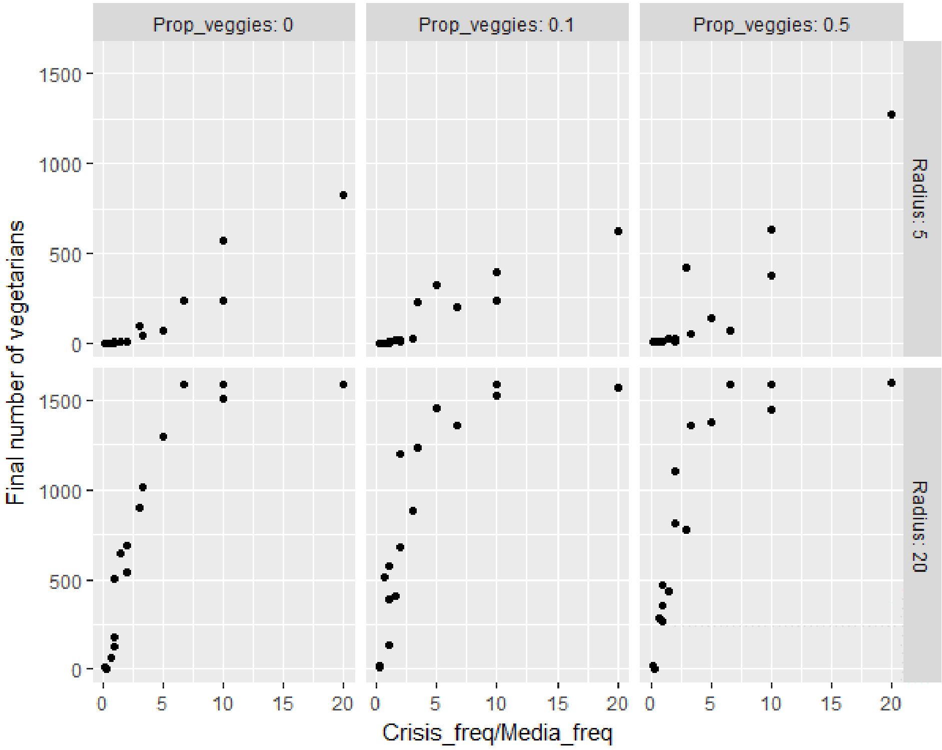

Figure 7: Combined effects of the initial proportion of vegetarians and of the radius of crises

Each step was been calibrated to correspond to about one month, so that the whole simulation corresponded to approximately 15 to 20 years.

Sensitivity Analysis

Sensitivity analysis raw results are given in Figures 6 to 9 and further commented in Section 3.3. The parameters analyzed are, respectively:

- the ratio between the frequencies of crises and communication campaigns (Figure 6);
- the initial proportion of vegetarians (Figure 7);
- the radius of crises (Figure 7);
- the progression over time of the resistance rate, constant or adaptive (Figure 8). In the latter case, the resistance rate tended to increase for individuals who had already experienced a food diet change, reflecting some "no going back" effect;
- the introduction of resistance to communication campaigns (Figure 9);
- the introduction of a selective influence of neighbours, where those with perceptions closer to the agent's perception are more carefully listened to (Figure 9).

3.3 Discussion

Our argument network structure revealed two main elements in particular. First, it was observed that arguments about Health were by far the majority of the arguments identified. They represented 47% of all of the 114 arguments identified and 63% of the arguments involved in at least one attack. As a matter of comparison,

Table 2: Comparison of argumentation and ABM models in this study.

	In argumentation	In ABM
Modelling scale:	The argument	The individual
Values (regarding food diet)	Are represented as criteria of the arguments (health, etc.)	Are synthetized by a level of needs for each individual
Assessment (of food diet)	Are represented by the type (+ or -) of each argument	Are expressed by perception levels that vary over time
Influences	Arguments may attack each other and all arguments may influence public opinion	Triggers positively or negatively impact perception levels
Resulting decision	Is attached to each argument through its type. Viewpoints can be computed but currently not quantified (e.g., through proportions).	Is attached to each individual through behavior (food diet). Trends for the whole population emerge from individuals.

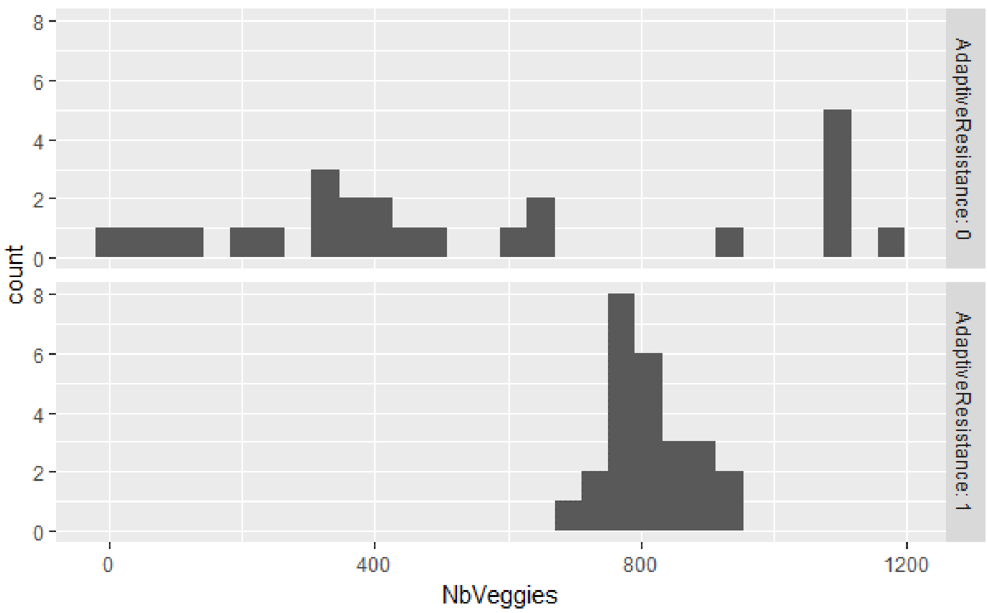

Figure 8: Effect of the adaptive resistance rate

ethical arguments represent only 3% of all arguments identified. Second, some key arguments emerged due to their centrality. Three arguments were involved in more than four attacks. The first argument, grouped under identification numbers 28, 57, 108 and 111 (the black node in Figure 2), refered to a scientific paper from the American Dietetic Association stating that "Properly planned vegetarian or vegan diets fit all stages of life" (Craig & Mangels, 2009). The second argument, grouped under identification numbers 71 and 72, was a journalist's statement that "No study is favourable to the vegan diet". The third argument, number 83, stated that "vegan diet safety is not proven", questioning the validity of scientific studies in this regard. These arguments would probably be key arguments in potential controversies about vegetarian diets due to their generality and to their polarized nature on the question of such diet viability from a health perspective.

The major importance of health issues surrounding vegetarian diets was in line with findings in Ruby (2012) review of vegetarian studies. In contrast the importance of ethical arguments that was stressed by Ruby (2012) did not appear in this modelling. This could be explained by the more complex nature of ethical arguments as well as our choice of search words in Google that focused on diets. However, from an ethical perspective, it seemed that the health issue (whether or not vegetarian diets are healthy) was central since vegetarianism may be defended from the baseline of animal products not being necessary for human health (Francione, 2015).

In this study, we built the network and proposed a structural analysis. Abstract argumentation led to further analysis and, in particular, the rejection of attacked arguments without any argument to defend them. Such analyses allow for new indicators such as those that measure the degree of controversy based on rejected argument ratios (Thomopoulos, 2018; Thomopoulos et al., 2017), which can better identify potential controversies.

The present work illustrates a promising coupling of the argumentation theory and agent-based modeling. The use of ABM is complementary since it allows for the exploration of conditions under which arguments could spread in a pop-

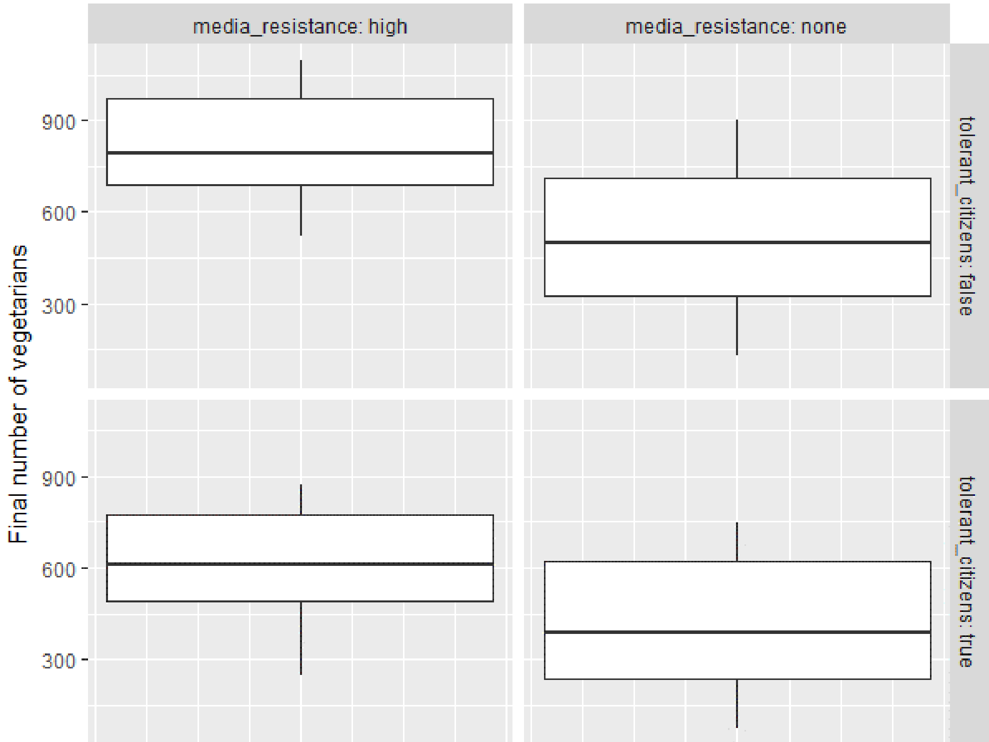

Figure 9: Effects of resistance to communication campaigns and of the selective influence of neighbours

ulation and favour the adoption of a vegetarian diet. The way arguments build public opinion and may lead to behavioural changes can be simulated by the ABM, whereas this dynamic cannot be captured by the argumentation system alone. A comparison of the argumentation and ABM representations that emerged from this study is proposed in Table 2.
The simulation results and, more specifically, sensitivity analysis, showed that arguments in favour of meat reduction can be widely diffused in the population and that the repeatability of this result is dependent on several parameters. Among them, the ratio between the frequencies of crises and communication campaigns plays an important part in the stability of the results obtained over the simulations (Figure 6). It is interesting to notice that the initial proportion of vegetarians has little effect compared to that of the radius of crisis impact (Figure 7). Moreover, another parameter was demonstrated to impact the stability of the model, namely the scalability of the resistance rate. Indeed, when resistance to change increases in the case of food diet change, the results in terms of the final number of vegetarians are much more homogeneous over the simulations (Figure 8). This observation is confirmed in the particular cases where this enhanced resistance affects consumer receptivity to communication campaigns and differently-thinking neighbours (Figure 9).
A crucial issue in argument modelling and simulation is the completeness and the robustness of the information collected. Of course, such a complex issue could never pretend to be exhaustive. So how can the question of completeness be dealt with? A first possible direction is to combine different types of information, in particular quantitative sources of information such as large surveys which allow analyzing the repres-

entativeness of different viewpoints, and qualitative ones which allow the identification of nuances. In this perspective, ongoing research extends the set of arguments elicited so as to include, on the one hand, quantitative results from a survey of 2,000 people questioned on their current and ideally intended food habits, and data from the French National Institute of Statistics and Economic Studies (INSEE) to model social networks at the level of a city; and on the other hand, qualitative results from 20 biographic interviews that provide in-depth understanding of the motivations, initiating events, brakes and facilitators of food diet changes on the long-term.
A second possible direction is to observe the evolution of the plus-value brought by new information sources in time. When no more substantial novelty is brought, a fixed point is reached.
Another thorny issue is the perceived reliability of arguments and its role in opinion dynamics. Indeed, the way arguments are processed can also be refined. Presently, argument perception depends on the criteria addressed by the argument and on the importance of these criteria for each citizen. Other factors can impact the perception of an argument, in particular, the source of the argument (Pornpitakpan, 2004). Hence, messages from dubious sources can differentially impact citizens' responsiveness. Various types of influencing agents can be distinguished, such as lobbies, the government, companies, etc., with various strategies to diffuse arguments.

4 Conclusions

The method presented here formalizes arguments and attacks around vegetarian diets using an abstract argumentation approach. The argument network revealed the foremost importance of health issues surrounding vegetarian diets. The centrality of some of the arguments in the network allowed for identification of potential key arguments and/or controversies. The importance of health arguments in relation to ethical arguments should be further researched.
Argument networks take advantage of being used together with ABM to explore the emergent establishment of new social norms in the case of vegetarianism. The overall research demonstrates the potential of developing an ABM to predict the triggers impacting the dynamics of food habit changes. Crises – sanitary, environmental and ethical – lead to the adoption of vegetarian products by consumers. The results indicated that there is the potential of extending the reach of nutrition transition by acting on several parameters: counterbalance of advertising campaigns by awareness campaigns, wide diffusion of awareness messages, and measures encouraging people to maintain their new habits and deterring them from backing down.
An interesting issue to further investigate is how public opinion is formed in the case of crises, including situations where the assumptions that the whole population is well informed and the news is reliable do not hold.

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Effect of Retort Processing on Low Sodium Instant Noodle Seasoning Based on Oil-in-Water Emulsions

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Abstract

Increasing consumption of instant noodles with high sodium content could elevate the risk of cardiovascular disease. Making instant noodle seasoning in the form of an oil-in-water emulsion was expected to improve the perception of salty taste without increasing the use of salt. However, the oil concentration in the emulsion affects the perception. The addition of antioxidant and retort processing was needed to overcome the nature of the emulsion that was quite susceptible to oxidation and microbial contamination. Preliminary research determined the optimum concentration of oil and antioxidant based on physical characteristics, the perception/gustation of saltiness, and emulsion oxidative stability and the results were used for further research. The final part of the research determined the effects of retort processing on emulsion stability, the perception of saltiness, and the degree of microbial contamination. Three different oil concentrations (26, 27, and 28%) and three types of antioxidants (natural vitamin E, ascorbyl palmitate and mixed tocopherol) were applied to the instant noodle seasoning oil-in-water emulsions. The results showed that using 28% oil and mixed tocopherol had the most stability, was more viscous, and had optimum salty taste perception, which significantly extended the shelf-life of the emulsion compared with the others. Retort processing for 21.5 minutes in 123.5^{o}C was applied to the seasoning emulsion with 28% oil content and mixed tocopherol. Although, the microbial contamination was significantly reduced, neither the stability of emulsion nor the perception of salty taste was significantly changed by the process.

Keywords: Instant noodle; O/W emulsion; Retort; Salt reduction; Seasoning

1 Introduction

The consumption rate of instant noodles is increasing globally, including in Indonesia. There is an estimated 1950 mg of sodium in one serving size of 120 g instant noodle, which exceeds the Recommended Dietary Allowance for common sodium intake of 1500 mg per day (BPOM, 2016; Daniel, Momoh, Friday, Okpachi, & Ejembi, 2014). High consumption of instant noodles can raise cardiometabolic risk which may lead to cardiovascular disease (Shin et al., 2014). Thus, a reduction in sodium intake is recommended to reduce and control blood pressure to avoid the risk of cardiovascular disease, which is the leading cause of death. One of the ways to reduce these risks is to make low sodium instant noodle seasoning in the form of an oil-in-water emulsion. Using an oil-in-water emulsion can increase the perception of saltiness in foods without in-

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creasing the use of NaCl (Torrico & Prinyawiwatkul, 2015). The presence of fat in the emulsion, which contributes to the mouthfeel, would tighten the binding of substrate (Na^{+}) with receptors in taste buds and enhance neurotransmitter transport, resulting in the transfer of the neuron electric signal to brain as salty perception. However, instant noodle seasoning emulsion tends to have low stability and poor sensory characteristics. Hence, instant noodle seasoning in the form of an emulsion is rarely found in the market (Bakry et al., 2015). The oil used in the emulsion is susceptible to oxidation, which would decrease the quality of the product due to the rancidity that occurs (Wasowicz et al., 2004). Therefore, the addition of antioxidant would prevent the rancidity. Moreover, the high-water content in the emulsion also leads to the proliferation of microorganisms, such as bacteria, yeasts and molds, and can also trigger deterioration due to oxidation (Lund, Baird-Parker, Baird-Parker, Gould, & Gould, 2000). Retort processing is applied to many food products to ensure their safety from microorganism contamination (Rahman, 2013) and instant noodle seasoning could be further improved using retort processing. The preliminary aim of this research was to obtain the optimum concentration of oil and antioxidant based on physical characteristic, the perception/gustation of saltiness, and emulsion oxidative stability and the results of which were used in the main research. The main aim was to determine the effects of retort processing on emulsion stability, the perception of saltiness, and the degree of microbial contamination in the emulsion.

2 Materials and Methods

Materials used were palm oil, instant noodle seasoning premix powder (reduced sodium 25%), control instant noodle seasoning premix powder, instant noodle, sweet soy sauce, xanthan gum, soy protein isolate (SPI), natural vitamin E, ascorbyl palmitate, mixed tocopherol, water, standard plate count agar, yeast extract glucose chloramphenicol agar, tap water, standard potassium solution 1000 ppm, standard sodium solution 400 ppm, phosphate buffer, alcohol, retortable pouches, and filter paper. Emulsions were prepared using a Silverson L5T High Shear mixer (Silverson, USA). Viscosity was measured using a Brookfield DV-E viscometer.

2.1 Preliminary Research

Emulsion Preparation and Emulsification Process

First, the aqueous phase was prepared using a constant concentration of premix seasoning powder, sweet soy sauce, SPI, xanthan gum, and antioxidant. The oil phase was prepared by weighing palm oil to 26, 27, and 28% of the total weight of the emulsion; the water used was calculated based on the oil concentration. The process of making instant noodle seasoning emulsion was done according to the method described by Arnold, Teja, and Yudianto (2018) with modification. The premix (sweet soy sauce 16.67% (w/w), premix seasoning powder 26.19% (w/w), SPI 1% (w/w), xanthan gum 0.1% (w/w) from aqueous phase, oil (26, 27, 28%) (w/w), antioxidant (natural vitamin E, ascorbyl palmitate, mixed tocopherol) 10 ppm and water (w/w)) were dry-mixed, while distilled water was heated to 70^{o}C. Then, soy sauce and the solid mixture were slowly poured into the water. The result of this mixing was referred to as liquid phase. Separately, the oil was mixed with the antioxidant to be used. Then the liquid and the oil mixture was homogenized using the high shear mixer for 10 minutes at 4000 rpm.

The emulsion produced was analyzed further for its sensory properties (saltiness evaluation). Sensory evaluation was carried out by thirty trained panellists on the same day as the emulsion sample production in the form of evaluating the saltiness perception of sample with scale test. The sample presented was in the form of a mixture of seasoned emulsion and instant noodles that had been boiled for the evaluation of saltiness. Panellists scores the perceived saltiness on a scale ranging from zero (very unsalty) to 15 (very salty). Viscosity was measured by Brookfield DV-E Viscometer, and emulsion stability was measured by creaming at 0, 7, 14, 21 and 28 days. Estimating shelf-life was determined by theAccelerated

shelf-life Testing (ASLT) method with the Arrhenius equation. The stability of the product was observed based on its critical parameters in the form of rancidity intensity during storage which was affected by the increase in temperature (Kilcast & Subramaniam, 2000).
The shelf-life of the sample was determined based on critical parameters in the form of sample oxidation level which is indicated by the rancidity value and expressed as separation fraction and for oxidation stability/shelf-life evaluation. Samples were stored at three different temperatures, namely 27^{o}C, 37^{o}C and 47^{o}C for 28 days, and the value of rancidity was observed on days 0, 7.14, 21, and 28. The results plotted to determine shelf-life of the product at a certain temperature, using the following equation:

$$K = K_{\infty} \cdot e^{\frac{-Ea}{RT}} \tag{1}$$

2.2 Main Research

Retort Processing

Retorting was carried out based on the method performed by Chung, Cha, Koo, Ahn, and Choi (1991) with modifications. Liquid phase and the oil mixture was homogenized using high shear mixer for 10 minutes at 4000 rpm. A total of 20 grams of seasoning emulsion (sweet soy sauce 16.67% (w/w); premix seasoning powder 26.19% (w/w); SPI 1% (w/w); xanthan gum 0.1% (w/w) from aqueous phase; oil (w/w), antioxidant 10 ppm and water (w/w)) were put into retortable pouches with dimensions of 9 × 15 cm. Retorting was carried out by autoclaving at 123.5^{o}C for 21.5 minutes. Each retort process was carried out on 32 retortable pouches simultaneously. After the retort process was complete, the effects of the processing on the instant noodle seasoning emulsion were measured by saltiness evaluation, emulsion stability, the degree of microbial contamination by total plate count (TPC)and yeast and mold count (YMC), moisture and sodium content. The analysis was also carried out on the non-retorted seasoning emulsion as a comparative treatment.

2.3 Replicates and statistical analysis

The data were analyzed using software IBM SPSS Statistics 22, with replication of three times per treatment with pairing for the preliminary research (ANOVA), and using the independent samples T-test of two treatments with four replications with each pairing for the main research.
Statistical analysis of the sensory properties and the effect of retort process carried out by Analysis of Variance (*one-way* ANOVA). All values were stated as the mean ± SD and statistical significance at $\alpha = 0.05$

3 Results and discussion

3.1 Preliminary Research

In the preliminary research, there were nine formulations which consisted of a combination of three types of antioxidant and three oil concentrations. Nine formulations of seasoning emulsion were a combination of 26% to 27% and 28 % oil (designated A, B, and C respectively), and the natural antioxidants vitamin E, ascorbyl palmitate and mixed tocopherols indicated by 1, 2, and 3. Thus the experimental treatments were designated A-1, A-2, A-3, B-1, B-2, B-3, C-1, C-2 and C-3 respectively. Analysis of the physical characteristics of the emulsion in the form of stability and viscosity, sensory saltiness and shelf-life of the emulsion based on the stability of oxidation was done to determine the best formulation to be used in the main study stage.

Viscosity

Based on the statistical analysis, the viscosity of emulsions was significantly affected by oil concentration, whereas type of antioxidant and the interaction between oil concentration and type of antioxidant were not significantly affected the viscosity of emulsions (α=0.05). Viscosity of emulsions with different oil concentration is shown in Table 1. It was discovered that the emulsion with 28% oil concentration had the highest viscosity, while emulsions with 26% oil concentration had the lowest viscosity (α=0.05). The viscosity of the emulsion was directly proportional to the increase in oil concentration. Higher oil concentration would increase the total of dispersed phase in emulsion, so that the higher density of the emulsion droplets, due to the higher oil concentration, would reduce the flow rate of the emulsion.

Table 1: Emulsion viscosity with different oil concentration

Oil concentration	Viscosity (cP)
26%	972.50±3.60^a
27%	986.33±3.31^b
28%	1067.89±2.61^c

Means with different superscript letter were significantly differenct (α=0.05)

Emulsion Stability

A higher separation fraction means the emulsions are less stable. After 28 days of observation, sign of declining emulsion stability was visible through the separation fraction. However, based on statistical analysis, the stability of emulsions was significantly affected by oil concentration, whereas type of antioxidant and the interaction between oil concentration and type of antioxidant did not significantly affect the stability of emulsions (α=0.05). Stability of emulsions with different oil concentrations is shown in Table 2. The emulsions with 28% oil concentration had the highest stability, whereas emulsions with 26% oil concentration had the lowest stability (α=0.05).
Emulsion stability increased with increased concentration of oil in the emulsion. According to Lin (2009), an emulsion containing 70% milk fat is relatively stable compared with 30% and 50% milk fat because of the static state of the droplets due to their density within the system such that the frequency of movement of the droplets that triggers separation decreases.

Table 2: Separation fraction of emulsions with different oil concentration

Oil concentration	Separation fraction (%)
26%	8.82±0.54^a
27%	7.95±0.63^b
28%	7.22±0.53^c

Means with different superscript letter were significantly differenct (α=0.05)

Perceived Saltiness of Emulsions

The intensity of the seasoning emulsion saltiness with different oil concentrations and type of antioxidant was evaluated using a line scale test.
Based on the statistical analysis, the perceived saltiness of the emulsions was significantly affected by oil concentration, whereas the type of antioxidant and the interaction between oil concentration and type of antioxidant did not significantly affect the stability of emulsions (α=0.05). Perceived saltiness of the emulsions with different oil concentration is shown in Table 3. Based on the post hoc test conducted on the effect of the concentration of oil, it was found that the emulsion with 28% oil concentration had the highest perception of saltiness. Furthermore, evaluation of perceived saltiness of the seasoning emulsions with three different types of oil concentration (A, B, C) and seasoning control (D) were conducted without the use of antioxidants. Sodium in seasoning powder used in the emulsion was reduced by 25% compared with the control.
Based on statistical analysis of the results in Table 4, there was a significant difference between the emulsions and the control. The emulsion with 28% oil concentration had the highest perceived saltiness, which was the same as the control seasoning. This indicated that the formation of oil in water emulsion with 28% oil concentration could reduce the use of sodium in the instant noodle seasoning by 25% without giving a different perceived saltiness. The presence of oil could accelerate the distribution of stimulus in the form of Na^+ ions by opening ion channel K^+ on the taste buds of the tongue and envelope the tongue. This may reduce the flushing process of the liq-

uid phase at the surface of the tongue so that the intensity of the saltiness increases (Stevens, Pashler, & Yantis, 2002; Suzuki, Zhong, Lee, & Martini, 2014). Increased oil concentration might also improve the concentration of tastants in the liquid phase in the emulsion. Thus, with the same amount of salt, the amount of dissolved salts will be more concentrated in the mouth when the liquid phase (water) in the emulsion is reduced. According to Torrico and Prinyawiwatkul (2015), the increase in concentration of oil in oil in water emulsion might increase the salty taste of the salt in certain concentrations.

Table 3: Line scale test of seasoning emulsions with different oil concentration

Oil concentration	Sensory score
26%	5.93±0.50-^{a}
27%	6.39±0.46^{b}
28%	6.77±0.56^{c}

Scale: 0= very not salty; 15= very salty; means with different superscript letter were significantly different (α=0.05)

Table 4: Line scale test of seasoning emulsions with different oil concentration and seasoning control

Formulation	Sensory score
A	5.98±0.43^{a}
B	6.32±0.42^{b}
C	6.86±0.47^{c}
D	6.79±0.43^{c}

Scale: 0= very not salty; 15= very salty; means with different superscript letter were significantly different (α=0.05)
A = Emulsion with 26% oil; B = Emulsion with 27% oil; C = Emulsion with 28% oil; D = Seasoning powder

Oxidative Stability

Oxidative stability was observed as shelf-life of the emulsion. The longer shelf-life, the higher oxidative stability of the emulsion. Based on the statistical analysis, the oxidative stability was significantly affected by oil concentration and type of antioxidant individually, whereas the interaction between oil concentration and types of antioxidant did not significantly affect the oxidative stability of the emulsions (α=0.05) (Table 5). Emulsions with 28% oil concentration had the longest shelf-life compared with the others, moreover the use of either ascorbyl palmitate or mixed tocopherol as antioxidant had a longer shelf-life than the use of natural vitamin E.

Emulsions with 28% oil concentration had the longest shelf-life which indicated that they had the best oxidative stability. The higher oil concentration increased the dispersed phase in the emulsion so increasing its viscosity and density of droplets in emulsion. Dense spacing reduces space between droplets in the emulsion so that air, as well as other components that act as precursors of oxidation are unable to enter the system and cannot contact the oil in the emulsion. According to Akoh (2005), decreasing the concentration of oil in the sample of oil in water emulsion from 30% to 10%, causes an increase in the total oxidative value, based on the peroxide and anisidine values of emulsion samples. At increased concentration of oil, the proportion of water fraction decreases with the increase in the oil fraction of the emulsion. As the concentration of water decreases, the soluble prooxidant in water would decrease thus decreasing the amount of free radical generation per droplet. High oil concentration increases the concentration of droplets of oil so that the total generation of free radicals will be decreased when compared with the total number of droplets. Fewer free radical would decrease the rate of oxidation in emulsion thereby increasing emulsion stability against oxidation. According to Mcclements and Decker (2009), when the oil droplet concentration is increased, there is decline in oxidized oil fractions from 43.75% to 6.25% in safflower oil in water emulsion containing sucrose.

Oxidative stability of emulsions with different types of antioxidant is shown in Table 6. Emulsions with natural vitamin E as an antioxidant had the shortest shelf-life, showing that vitamin E was less effective in promoting oxidative stability. The low effectiveness of vitamin E could have occured due to the addition of natural vitamin E which increased the amount of natural tocopherol in oil which resulting prooxidative effect. The addition of natural vitamin E as an antioxidant could also cause antagonistic interactions with natural antioxidants in the oil. According to Choe and Min (2009), there is an antagonistic interaction between the plant extract rich in polyphenols with α-tocopherol in pork fat or sunflower seed oil. It could also be caused due to the fact that natural vitamin E is less stable than the other two types of antioxidants that were used. The use of mixed tocopherols and ascorbyl palmitate as an antioxidant in the seasoning promoted significantly longer shelf-life. This indicated that both types of antioxidants were good at maintaining the stability of the emulsion instant noodle seasoning against oxidation. Ascorbyl palmitate effectively works as an antioxidant in vegetable oils either alone or in combination with other natural antioxidants. According Shahidi (2015), ascorbyl palmitate synergistically prevents the oxidative reaction with α-tocopherol, which is naturally present in corn oil. The tocopherol consists of a mixture of the tocopherol derivatives α-, β-, γ-, δ- tocopherol which have antioxidant effects that are influenced by the composition of the mixture. A mixture of oil-soluble tocopherol produces the best protection at a concentration of 16 ppm in mayonnaise, whereas the antioxidant effect is reduced when the concentration is too high or too low.

Table 5: shelf-life of emulsions with different oil concentration

Oil concentration	**shelf-life (days)**
26%	264.45 ± 7.54^a
27%	298.52 ± 8.16^b
28%	322.33 ± 4.68^c

Means with different superscript letter were significantly differenct (α=0.05)

3.2 Main Research

Main research aimed to determine the effect of retort processing by using emulsion with 28% oil concentration and the antioxidant chosen from preliminary research, tocopherol. Emulsions were retorted and then compared with a control group without retorting. The emulsions were analyzed for their stability, microbial contamination, and perceived saltiness.

Table 6: shelf-life of emulsions with different types of antioxidant

Antioxidant	**shelf-life (days)**
Natural vitamin E	28.88 ± 30.09^a
Ascorbyl palmitate	297.62 ± 23.70^b
Mixed tocopherol	298.79 ± 25.26^c

Means with different superscript letter were significantly differenct (α=0.05)

Emulsion Stability

Emulsion stability was evaluated by measuring the level of creaming that occurred on day 0, 7, 14, 21 and 28 based on separation fractions (Figure 1). The higher the separation fraction the less stable the emulsions. Based on the observation for 28 days, it was known that the separation of the emulsion occured and increased during the observation period for both emulsions. Statistical analysis showed that there was no significant difference in the stability between emulsion with or without retort processing (α=0.05) (Figure 2). Both emulsions appeared to have an increased level of separation with the same trend until the end of the observation period. These results may have been obtained due to protein changes in the emulsion during the retorting process. According to Kerry (2012), a thermal process such as retorting could change the structure of secondary, tertiary and quaternary protein which could also break the bonds of the protein so that the protein structure is open. Protein functional and physiological changes could occur due to chemical modification of the amino acid or from protein denaturation during the application of heat.
The heating process affects emulsifier adsorbing at the oil droplet surface. When the absorbed globular protein is heated above the temperature of denaturation, the structure and chemi-

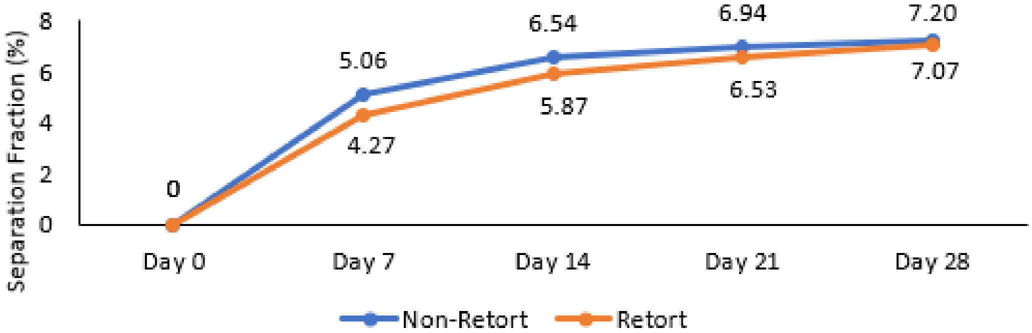

Figure 1: Separation fraction of emulsion with and without retort processing

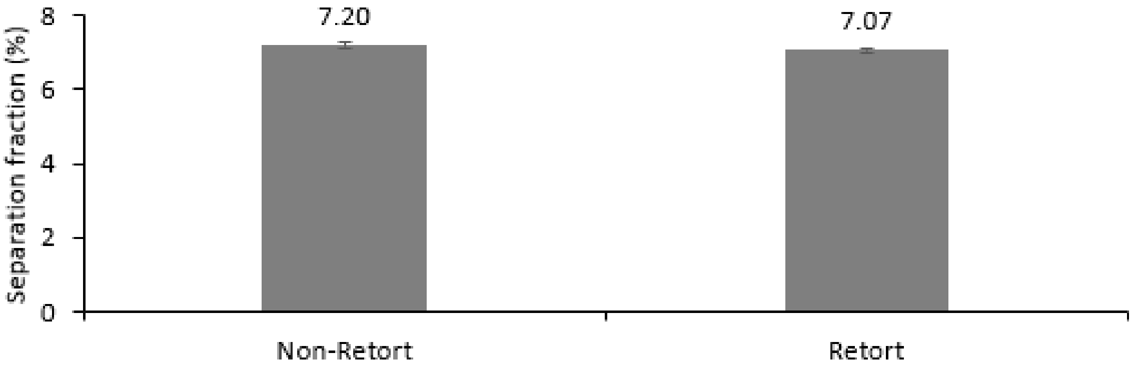

Figure 2: Separation fraction of emulsion with and without retort processing on day 28

Table 7: TPC and YMC of emulsion with and without retort processing

	Non-Retort			Retort		
	TPC (CFU)	Yeast (CFU)	Mold (CFU)	TPC (CFU)	Yeast (CFU)	Mold (CFU)
Day 0	1.74 x 10^{3a}	0	0	0^b	0	0
Day 7	5.26 x 10^{3a}	0	0	0^b	0	0

CFU – colony forming units. Means different superscript letter on the same row (α=0.05)

Table 8: Sensory saltiness scores, moisture, and sodium content of emulsion with and without retort processing

	Non-Retort	Retort
Saltiness	4.70±1.02^a	5.17±1.37^a
Moisture Content %	33.62 ± 0.19^a	34.78 ± 0.27^a
Sodium (mg/100g)	2000.23 ± 120.68^a	1902.17 ± 104.26^a

Saltiness scale: 1= very not salty; 7= very salty; means with different superscript letter on the same row (α=0.05)

cal activity of proteins changes irreversible. Proteins unfold completely and could effectively rearrange all nonpolar amino acid in the oil phase, which decreases the effect of aggregation. At higher temperatures, protein in the interphase membrane would make a more compact layer in the surrounding oil droplet. It would increase droplet density and decrease the probability of creaming. In milk heated between temperatures of 65-80^{o}C that the fat droplet size is enlarged by partially denaturing the protein thereby increasing the rate of droplet flocculation and the surface of the droplet becomes more hydrophobic, whereas at higher temperatures (80-90^{o}C), the size of the droplets become smaller and compact than before (Raikos, 2010). Additionally, protein changes could decrease protein component solubility so the viscosity would increase as well, which stabilizes the emulsion from creaming. According to Liang (2014), the changing in temperature can result in changes to the protein components occuring, such as aggregation, dephosphorization, deamination, polymerization and hydrolysis, all of which affect the nature of the protein component. Crosslinking may occur between the amino acids of protein that can reduce protein solubility.

Complete of partial protein denaturation may involve folding that causes aggregation. Increased in pressure homogenization or the application of heat at a temperature of 90^{o}C resulted in denaturation of proteins causing the formation of aggregate particles which increased the viscosity and gelation of the emulsion system (Buxbaum, 2007).

Microbial Contamination

Total plate count and yeast mold count from the emulsions with and without retorting are shown in Table 7. It was known that microbial contamination increased significantly in the emulsion without retort processing. The retort treatment showed a significant reduction in microbial contamination as seen in total plate count (α=0.05). Retort treatment could produce sterile conditions in the emulsion and maintain that condition until the seventh day of observation. High temperature treatment at 123.5^{o}C for 21.5 minutes served as the sterilization process for the emulsion so there was no microbial contamination in the product (Zeuthen & Bøgh-Sørensen, 2003).

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Perceived Saltiness of Emulsion

Scoring test of perceived saltiness showed that there was no significant difference between emulsion with and without retort processing (α=0.05) (Table 8). The retort process did not affect the intensity of perceived saltiness in seasoning emulsion. The emulsion was stable to heat treatment, so it was not broken and therefore could maintain the function of oil-in-water emulsion that can increase the perception of saltiness. This result was also supported by the moisture and sodium contents that were not significantly different between the seasoning emulsion with and without retort processing (Table 8). It showed that the retort process would still maintain the water concentration within the emulsion as it did not cause evaporation. The retortable pouch consists of several layers based on a layer of aluminum foil that acts as an absolute barrier to mass transfer, light and microorganisms (Shihab, Hafeeda, Kumar, Tamilselvan, & Nadanasabapathi, 2013), making it impermeable to gas and water. Hence, it could prevent the transmission of water vapour and thus the moisture content of products does not change significantly. The sodium content of seasoning emulsion with and without retort processing was not significantly different (Table 8). Heat treatment such as retorting at 123.5^{o}C for 21.5 minutes had no significant effect on the sodium content of the instant noodle seasoning emulsion. Thus, the intensity of the salty taste between emulsions was not significantly different.

4 Conclusions

Based on this research, the instant noodle seasoning emulsion with 28% oil concentration had the highest oxidative stability, stability, and viscosity as well as the highest level of saltiness which was not significantly different from the control seasoning powder without sodium reduction. Moreover, the use of mixed tocopherol as antioxidant in the emulsion produced the highest oxidation stability. Thus, the combination of 28% oil concentration and mixed tocopherol was the best formulation and used for retort processing. This research showed that retort processing had no significantly effect on the stability of emulsion and the intensity of saltiness in instant noodle seasoning emulsion. However, retort processing did significantly decrease microbial contamination in instant noodle seasoning emulsion based on the total plate counts. Further research is still needed on the application of oil in water emulsion on a low-sodium seasoning instant noodles with the retort process. The influence of retorting on the reduction of sodium in the instant noodle seasoning should be investigated. Further evaluation regarding colour and flavour with the retorting process is required to determine consumer acceptance of the product. In addition, to improve energy and cost efficiency in this application, it is necessary to optimize the treatment retorting to the instant noodle seasoning emulsion. Evaluation of shelf-life to rancidity and flavour acceptance and specific microbial contamination should be done to assess the effectiveness and efficiency of the retort treatment against the commercial standard specification instant noodle seasoning. This would make the expected results more applicable to the instant noodle seasoning emulsion.

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Thermal Degradation of β-Carotene from Macauba Palm: Mathematical Modeling and Parameter Estimation

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Abstract

Worldwide, there is a current need for new sources of vegetable oils. The natural content of total carotenoids in *Acrocomia aculeata* palm oil (up to 378 μg.g^{-1}) surpasses that of many other tropical fruits, making it one of its main compositional characteristics. As far as can be verified, there is no available information on the degradation kinetics of carotenoids for *A. aculeata* oil, which is required to describe reaction rates and to predict changes that can occur during food processing. The present study considered prediction abilities that have emerged with the use of specific kinetic data and procedures to understand thermal processing better, as an essential unity operation. Two kinetic mechanisms were proposed to describe the overall thermal degradation of carotenoids in the oil; the first one consists of three reaction steps while the other presents only one-step reaction. Mass balance equations were numerically solved by a Backward Differentiation Formula technique. The kinetic parameters from both models were estimated through a hybrid optimisation method using the Particle Swarm Optimization and the Gauss-Newton method, followed by statistical analyses. The model with more than one reaction was shown to be overparameterized and was discarded. The model with a single reaction was highly suited to handle the experimental data available, and the dependency of its rate constant on temperature was expressed according to Arrhenius law. As far we know, this is the first time the kinetics of carotenoids thermal degradation in *A. aculeata* oil is investigated through modelling simulation.

Keywords: Parameter Estimation; β-Carotene; Mathematical Modeling

1 Introduction

From the perspective of extractive practices concerning oil crops, *Acrocomia aculeata*, known as macauba, is a palm tree widely dispersed in Brazil. Over the last few years, the food and chemical industries have been responsible for most of the growing demand for vegetable oils. In this context, the interest in macauba as a food product has increased due to factors such as the nutritional quality of the oils extracted from its edible parts. The industrial interest in macauba has also involved full use of its fruit to generate co-products with added value. The fruit

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mesocarp and kernel together correspond to approximately 47% (on dry basis) of the total fruit weight. Noticeably, the mesocarp contributes to around 60% (on dry basis) of the total oil content, with a predominance of oleic ω-9 (53%) and linoleic ω-6 (18%) acids. The kernel oil is predominantly saturated, with around 40% of lauric acid.

An adult *Acrocomia aculeata* palm fructifies almost throughout the year with productivity ranging from 4 to 6 tonnes of esculent oil per hectare. The crop has, accordingly, a similar productive potential to *Elaeis guineensis*, which is among the highest oil-yielding plants in the world (Evaristo et al., 2016). Considering that high yield has been an essential characteristic for the selection of commercial harvests, it has potential as an alternative oil crop that deserves further investigation (Rodriguez-Amaya, Kimura, Godoy & Amaya-Farfan, 2008). Regarding the oil extracted from the fruit mesocarp, it contains no antinutritional factors and contains up to 378 $mg.kg^{-1}$ of total carotenoids, mainly β-Carotene (Nunes, Favaro, Galvani & Miranda, 2015).

The carotenoids are essential pigments in fruits, these tetraterpenes (C40) synthesised by plants are secondary metabolites, necessary for photosynthesis and to prevent photo-oxidation induced by light intensities. These functions are a consequence of the light-absorbing properties of their polyene chromophore (Oloo, Shitandi, Mahungu, Malinga & Ogata, 2014; Rodriguez-Amaya, Rodriguez & Amaya-Farfan, 2006; Schieber & Carle, 2005). In general, carotenoids naturally exist as all-trans form. However, isomerisation of all-*trans*-carotenoids to cis forms is one of the major reactions of the compound's degradations. The critical step for losses of the component in vegetable oils remains related to the exposure to high temperature, light or pro-oxidant molecules. Indeed, the elevation of temperature during thermal treatments has been shown to dramatically increase corresponding degradation reactions rates (Achir, Randrianatoandro, Bohuon, Laffargue & Avallone, 2010; Sampaio et al., 2013).

The intensity of the thermal treatment is a critical factor that has to be controlled to increase carotenoid retention in the macauba mesocarp oil. As an essential unit operation, the thermal processing influences both carotenoids bioaccessibility and the health-related attributes of vegetable oils (Nunes et al., 2015; Palmero et al., 2013; Rodriguez-Amaya et al., 2006).

Along with carotenoids products' identification, kinetic data become necessary to predict carotene loss on thermal degradation accurately. Kinetic evaluation is therefore required to derive necessary kinetic information for a system to describe the reaction rate as a function of experimental variables also predicting changes in a particular food system during processing. In general, most of the studies in real food report a first-order reaction on the concentration of *trans*-β-carotene, in different systems, at different processing temperatures. Although zero-order equations have also been verified, the use of a first-order kinetic is realistic in most cases. As far as we have knowledge there is no available information about thermal degradation kinetic modelling of neither β-carotene nor any other carotenoids from macauba oil (Achir, Penicaud, Avallone & Bohuon, 2011; Ahmed, Shivhare & Sandhu, 2002; Knockaert et al., 2012; Penicaud, Achir, Dhuique-Mayer, Dornier & Bohuon, 2011).

Some studies in nonpolar solvents tested reaction orders superior to one for trans-β-carotene degradation and found a better fit of experimental data by linearisation or nonlinear regression methods. The superior orders may be explained by the competition with isomerisation reactions, which are also of importance in vegetable oils. Most of the kinetic models used to describe trans-β-carotene degradation are single response kinetic models. However, as the compound is supposed to generate various degradation products, the original reaction scheme can be complex to involve complex dynamics (Achir et al., 2010; Penicaud et al., 2011; Sampaio et al., 2013). Regarding the estimation of kinetic parameters for trans-β-carotene degradation, the rate constants k (s^{-1}) can vary ranging from 0.00018 (120 ^{o}C) to 0.0015 (180 ^{o}C). The apparent activation energy E_a ($kJ.mol^{-1}$) tends to range from 80 to 110 (Achir et al., 2010; Dhuique-Mayer et al., 2007; Henry, Catignani & Schwartz, 1998; Sampaio et al., 2013).

The objective of the present study was to predict the thermal degradation of all-*trans*-β-carotene in *Acrocomia aculeata* oil through mathematical modelling, emphasizing the importance of numerical predictions for practical applications. This study is an important step to comprehend better the kinetics of thermal degradation of macauba oil, which is a promising source of high-quality raw materials.

2 Materials and Methods

2.1 Crude Oil: *Acrocomia aculeata*

Acrocomia aculeata fruit was collected from native palms with a maximum of five days after the fall in the Federal University of Minas Gerais - UFMG, located in the metropolitan region of Belo Horizonte, Minas Gerais, Brazil. The mesocarp and kernel portions were promptly separated from the fruit. Before the oil extraction, the mesocarp and kernel portions were thawed, air-dried at 60 ^{o}C for 48 hours and comminuted in an electric grinder coupled to a stainless steel cup (Goula, 2013; Pimenta, 2010). For the present study, the samples consisted of edible oil mechanically obtained from the macauba mesocarp by a continuously operated Expeller® press, at 34 ^{o}C. Amber glass vials (15 mL) were filled to the maximum working volume with the samples, minimising the impact of light and oxygen intrusion by reducing the volume of headspace. Samples were stored at freezing temperature (18 ^{o}C) until the analysis to minimise possible rates and extents of enzymatic lipolysis, also concerning potential losses of antioxidants (Koidis & Boskou, 2015; Parducci & Fennema, 1978).

2.2 Carotenoid Determination

The HPLC analyses of carotenoids were carried out on a Shimadzu system (Shimadzu, Japan) equipped with a vacuum degasser, a quaternary pump and an autosampler (SIL-20A HT). A UV-Visible photodiode array detector (SPD-M20A) was set in the range of 190 – 800 nm to analyse the chromatograms. Once β-carotene represent around 90% of the total carotenoids content in the A. aculeata mesocarp oil (Coimbra & Jorge, 2012; Nunes et al., 2015), peaks were detected at 455 nm, and the results were expressed as all-*trans*-β-carotene. The separation was achieved at 30 ^{o}C using a normal phase column (Phenomenex Luna Silica (2) 100A Si: 250 mm $\times$ 4.6 mm i.d., 5 μm particle size) prior equilibrated with a flow of 0.1 mL.min^{-1}. The mobile phase was n-hexane/isopropyl alcohol (97.0:3.0 v/v), the flow rate was maintained at 1.0 mL.min^{-1}, and the elution remained isocratic until 26 min. After every 10 injections of 20μL, the column was reactivated with a solution of 10% isopropyl alcohol in n-hexane (v/v). The carotenoids were identified for each experimental condition by the combined use of their relative retention times and previously published UV/Vis spectra (Panfili, Fratianni & Irano, 2004; Rodriguez-Amaya, Kimura et al., 2004). The concentrations (mg.kg^{-1}) of carotenoids were expressed based on external matrix calibration (Sigma: St. Luis, MO). The spectrophotometric determination of total carotenoids (SD-TC) was performed using a Hach DR 2800 spectrophotometer (Hach, Loveland, CO, USA) as recommended by Rodriguez-Amaya, Kimura et al. (2004). The quantification (mg.kg-1) considered an absorption coefficient $A_{1\ cm}^{1\%}$ of 2580 in high purity n-hexane (Zscheile, White, Beadle & Roach, 1942).

2.3 Thermal Degradation of all-trans-β-Carotene

For all the experiments, the initial carotenoid concentration (determined according to item 2.2) was applied. 0.20 mL of oil (ρ-macauba oil = 925.6 g L^{-1}) was used. The thermal treatment that supported the Model Development and the Numerical Approach was carried out in triplicate, on a dry thermo-block (SAE 1020 steel Dry-Block - CE-350; Cienlab). Aliquots of oil (2000 μL) were placed into glass vials and inserted into the reactor. The aliquots were then heated until reaching the set temperature (t0). Thus, the heat treatments were assumed isothermal. The temperature monitoring (accuracy of $\pm$ 1 ^{o}C) was performed based on digital thermometer probes placed into three control vials, also containing 2000 μL of oil. The whole device was covered with aluminium foil to prevent carotenoid degradation by light. The thermal degradation kinetics was carried out employing at least five time sampling points, at five temperatures: For 100 ^{o}C, the experiment was run until 1500 min was reached; for 110 ^{o}C, 1440 min; for 130 ^{o}C, 270 min; for 140 ^{o}C, 160 min and, finally, for 150 ^{o}C, the experiment was conducted during 120 min.

2.4 Model Development and Numerical Approach

A first-order kinetic mechanism (Mechanism I) with three irreversible reaction steps, represented in Equations (1) to (3), was initially chosen to be tested if it can represent well the thermal degradation of β-carotene in macauba oil.

$$A \xrightarrow{k_{AB}} B \tag{1}$$

$$A \xrightarrow{k_{AC}} C \tag{2}$$

$$C \xrightarrow{k_{CB}} B \tag{3}$$

In the above equations, A represents the all-*trans*-β-carotene, B represents the oxidation and cleavage products (OCP), C represents *cis*-carotenoids (mainly, 9 or 13 -*cis*-β-carotene). Also, k_{AB} and k_{CB} correspond to the rate constants for the thermal degradation of all-*trans* and *cis*-β-carotene, respectively. k_{AC} corresponds to the carotene rate constant for the isomerisation of trans-β-carotene.
Assuming an isothermal, the material balances for all species involved in Mechanism I are represented by:

$$\frac{d\,[A]}{dt} = -k_{AB}\,[A] - k_{AC}\,[A] \tag{4}$$

$$\frac{d\,[B]}{dt} = k_{AB}\,[A] + k_{CB}\,[C] \tag{5}$$

$$\frac{d\,[C]}{dt} = k_{AC}\,[A] - k_{CB}\,[C] \tag{6}$$

A second mechanism (Mechanism II) was also considered to be a candidate mechanism able to predict well the experimental data available from the thermal degradation of β-carotene. This second mechanism is a simplified version of the first one, and it is shown in Equation (7).

$$A^{*} \xrightarrow{k_{A^{*}B}} B \tag{7}$$

In the above equation, A^{*} represents the sum of all-*trans* and *cis*-β-carotene, and k_{A*B} correspond to the rate constant for the thermal degradation of β-carotene.
Once more, assuming an isothermal, the material balances for all species involved in Mechanism II are represented by:

$$\frac{d\,[A^{*}]}{dt} = -k_{A^{*}B}\,[A^{*}] \tag{8}$$

$$\frac{d\,[B]}{dt} = k_{A^{*}B}\,[A^{*}] \tag{9}$$

The ordinary differential equations resultant from the material balances (Equations (4) to (6) and (8) and (9)) were then solved using the BDF (backward differentiation formula) technique, as programmed in the DASSL code (Petzold, 1982). Parameter estimation was performed with the package ESTIMA, implemented in Fortran, using a hybrid optimisation method PSO (particle swarm optimisation) and Gauss-Newton algorithms (Brandao, Oechsler, Gomes, Souza & Pinto, 2018; Schwaab, Biscaia, Monteiro & Pinto, 2008). Five hundred particles were used, and two thousand iterations were performed with a numerical tolerance of 0.0001 for the objective function. Besides, a confidence level of 95% was considered, and all the parameters from Mechanisms I and II were estimated in their absolute form. The known weighted least-squares function was used as the objective function in the present research, and it was defined as follows:

$$F_{obj} = (y^{e} - y^{m}(x^{m},\theta))^{T} V_y^{(}-1)(y^{e} - y^{m}(x^{m},\theta)) \tag{10}$$

In the above equation, y^{e} and y^{m} are the vectors for the measured and predicted dependent variables, respectively; V_y is the covariance matrix of the measured outputs (assumed to be diagonal); and x^{m} and υ are the vectors of the measured independent variables and model parameters, respectively.
Experimental variances were obtained through replicates and are illustrated in the following sections. The experimental data used to estimate the parameters was the amounts of all-*trans*-β-carotene concentration presented at certain times (A) divided by the initial amount of the caroten-

oid (A_o). Figure 1 shows the parameters estimation procedure performed in this study.

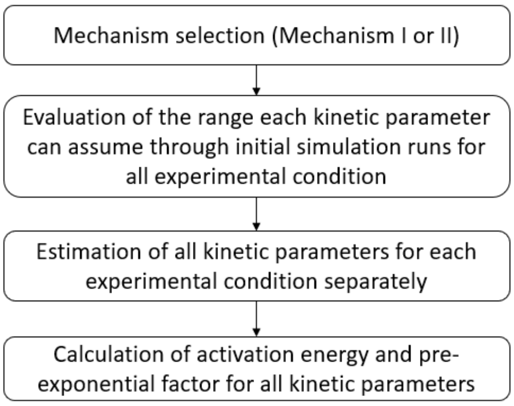

Figure 1: Flowchart of the parameter's estimation procedure

3 Results and Discussion

3.1 The task and project brief

The initial concentration of carotenoids was determined as 224.1 mg for each Kg of macauba oil. Regarding the estimation procedure, it was conducted for each experimental condition separately, as shown in Figure 1. Table 1 shows the results for the objective function, the estimated parameter and respective uncertainty obtained when model derived from Mechanism I was considered. The experimental data and the model predictions within their uncertainties are shown in Figure 2.

From Table 1, except for reaction temperatures of 140 and 150 ^oC, it is possible to observe that parameters uncertainties could not be calculated. Although all objective functions values were between the lower and upper limits of the $\chi 2$-distribution, the first model was not able to fit the experimental data available well since the model is over parametrised. At experimental conditions 140 and 150 ^oC, practically all parameters can assume zero value, which shows they are not significative and, consequently, they can receive any value that won't change model prediction including zero or even negative values. Figure 2 shows that predicted and experimental profiles for the ratio A/A_0 during the reaction are in good agreement which was expected since the model has more parameters it requires, so more than one parameter combination can provide apparent good results. Since the parameters uncertainties for experiments at temperatures 100, 110 and 130 ^oC could not be obtained through the parameter estimation procedure, as was done for the experiments done at 140 and 150 ^oC, at least the parameters confidence regions for these three experimental conditions could be determined, providing an idea about the values the parameters k_{AB} , k_{AC} and k_{CB} can assume. So, Figure 3 shows the parameters confidence regions for experiments conducted at 100, 110 and 130 ^oC using the model derived from Mechanism I. In all confidence regions the estimated parameter values, presented in Table 1, are highlighted.

From Figure 3, it is possible to observe that, in all confidence regions, at least one kinetic parameter can assume zero value or even negative values. The regions are not well defined since parameter uncertainties are significantly high, also suggesting that the confidence region of parameters uncertainties become open (Schwaab et al., 2008), indicating estimability problems. The estimation of all parameter uncertainties simultaneously is unfeasible for these experimental conditions using Mechanism I. Based on what was said, the model derived from Mechanism I can be unconsidered as a candidate model to represent the experimental data available of thermal degradation of macauba oil.

The final objective function values for the five estimations done were between the lower and upper limits of the $\chi 2$-distribution. Therefore, according to the $\chi 2$ statistical test, the proposed model fitted experimental data well, and was suitable to predict the experimental data available. Besides, all parameters uncertainties could be calculated, and they are smaller than the estimated parameters. Figures 4 shows that simulated and experimental trajectories for A/Ao during the reaction are in very good agreement for runs conducted at 100, 110, 130, 140 and 150 ^oC. The model predictions adequately fitted the experimental data measured during reactions, respect-

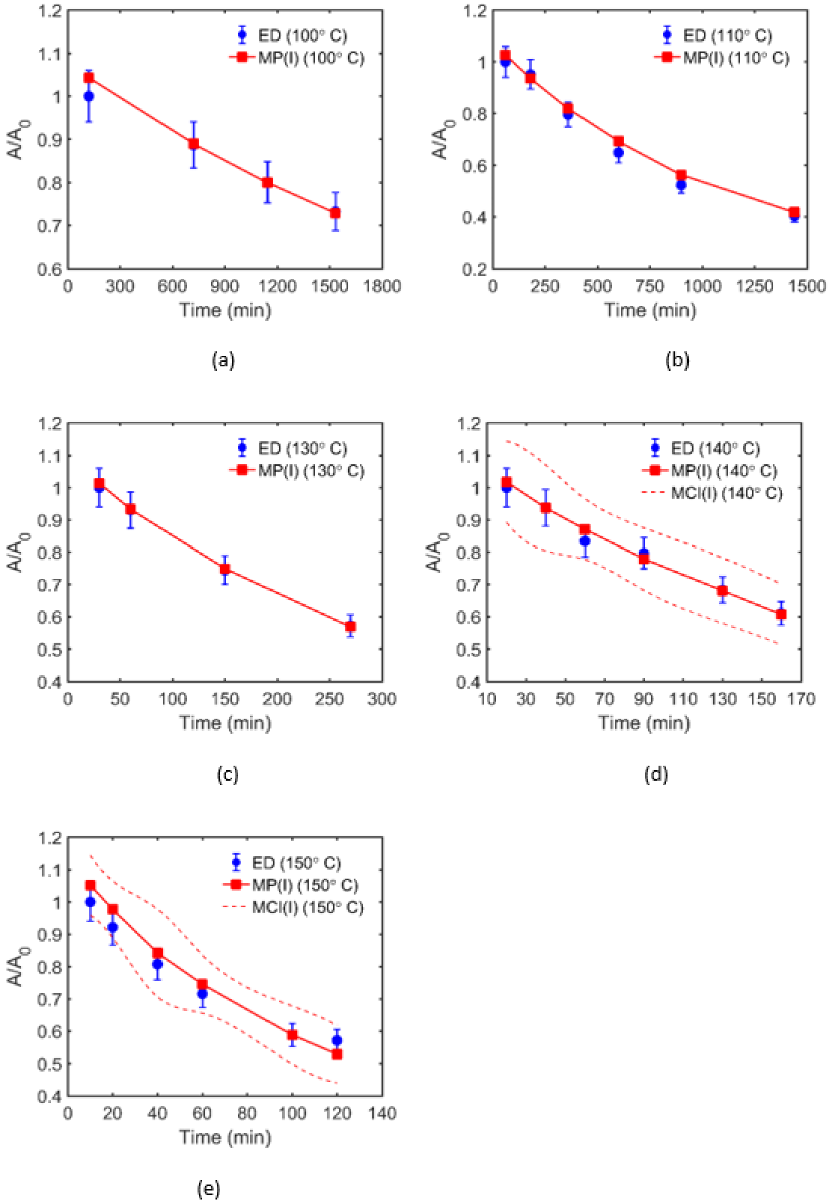

Figure 2: Predicted and experimental data at (a) 100, (b) 110, (c) 130, (d) 140 and (e) 150 ^{o}C using model derived from Mechanism I (MCI (I) = model confidence interval using Mechanism I; MP (I) = model predictions using Mechanism I; ED = experimental data).

Table 1: Parameter estimation results using Mechanism I[a]

T (^{o}C)	**k$_{AB}$ (min^{-1})**	**k$_{AC}$ (min^{-1})**	**k$_{CB}$ (min^{-1})**	**F$_{OBJ}$**	**X$^2_{INF}$**	**X$^2_{SUP}$**
100	$6.44 \cdot 10^{-9}$	$2.80 \cdot 10^{-4}$	0.0404	0.53	0.0009	5.02
110	$2.45 \cdot 10^{-4}$	$6.04 \cdot 10^{-4}$	0.00922	0.80	0.22	9.35
130	$3.23 \cdot 10^{-4}$	0.00245	0.067	0.08	0.0009	5.02
140	$1.64 \cdot 10^{-5} \pm 0.0017$	0.0041 ± 0.0109	0.114 ± 1.08	0.79	0.22	9.35
150	$1.30 \cdot 10^{-9} \pm 0.0019$	0.0074 ± 0.0054	0.0872 ± 3.23	4.24	0.22	9.35

a. (T = temperature, F$_{OBJ}$ = objective function, X$^2_{INF}$ = lower bound of chi-squared distribution, X$^2_{SUP}$ = upper bound of chi-squared distribution)

Table 2: Parameter estimation results using Mechanism II

T (^{o}C)	**k$_{AB}$ (min^{-1})**	**F$_{OBJ}$**	**X$^2_{INF}$**	**X$^2_{SUP}$**
100	$0.00025 \pm 1.4 \cdot 10^{-5}$	0.99	0.22	9.35
110	$0.00071 \pm 5.0 \cdot 10^{-5}$	9.70	0.83	12.83
130	$0.00246 \pm 4.1 \cdot 10^{-5}$	0.78	0.22	9.35
140	$0.00375 \pm 5.3 \cdot 10^{-4}$	1.87	0.83	12.83
150	$0.00681 \pm 1.2 \cdot 10^{-3}$	10.86	0.83	12.83

ing model and experimental uncertainties.
At this point, the remaining candidate model is only the model derived from Mechanism II. Table 2 presents the parameter estimation results using this model.
The temperature dependence on the reaction rate constant k_{A*B} is quantified by the activation energy Ea (KJ mol^{-1}) and is expressed by the Arrhenius equation represented in Equation (11). This effect of temperature on the kinetic constant k_{A*B} is shown in Figure 5. It can be observed that the estimated kinetic parameters follow the Arrhenius correlation very closely since the rate constants increase with the increase of reaction temperature. Linear regression, with a coefficient of determination (R^2) equals to 0.9891, was carried out to calculate the activation energy (E_a) and pre-exponential factor (A) for k_{A*B} constant shown in Table 2. The obtained fits can also be observed in Figure 5.

$$K_{A*B} = A.exp(-\frac{Ea}{RT}) \qquad (11)$$

In the presented Equation (11), T is the reaction temperature in Kelvin (K), A is the pre-exponential factor expressed in the same units as k_{A*B} and R is the universal gas constant (8.314 J mol^{-1} K^{-1}).

For total all-*trans*-β-carotene degradation, a pre-exponential factor of $1.58 \cdot 10^8$ min^{-1} and an activation energy of 83.8 kJ mol^{-1} K^{-1} were found. Knockaert et al. (2012) reported a rate constant at 110 ^{o}C of 0.10 ± 0.01 min^{-1} and an activation energy of total β-carotene degradation for both an olive oil/carrot emulsion of 45.0 ± 8.6 kJ.mol^{-1}. On the other hand, Chen and Huang (1998) reported an activation energy equal to 39 kJ/mol for total β-carotene degradation of an all-*trans*-β-carotene standard dissolved in hexane. Depending on the carotenoid source, as well as on the processing conditions and the reaction medium, a range from 20 to 171 kJ/mol has been reported for the activation energy related to the thermal degradation of β-carotene (Achir et al., 2011; Achir et al., 2010; Penicaud et al., 2011; Sampaio et al., 2013). To the best of our knowledge, the kinetics of the thermal degradation of β-carotene in *Acrocomia aculeata* oil has not been investigated yet. Therefore, this result shows the thermal degradation kinetics for the all-*trans*-β-carotene in the oil agrees with those of previous studies carried out on other diverse sources of vegetable oils, since the activation energy estimated in the present study is indeed

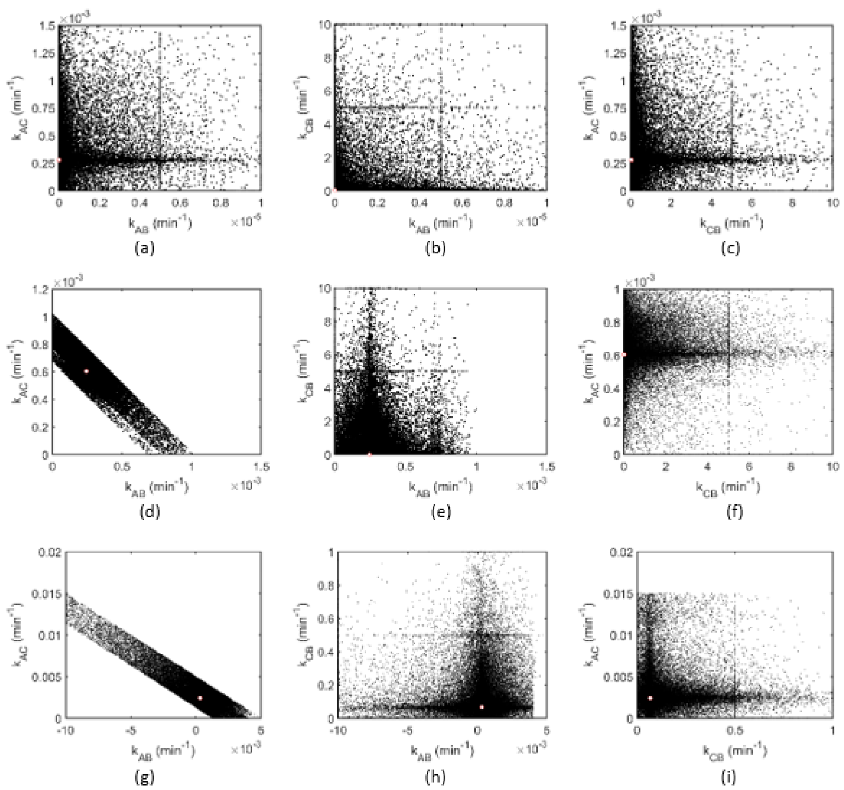

Figure 3: Parameters Confidence Regions, when Mechanism I was considered, at reaction temperature equals to (a), (b) and (c) 100 ^oC, (d), (e) and (f) 110 ^oC, (g), (h) and (i) 130 ^oC.

within the range reported by other authors.

4 Conclusion

The thermal degradation predicted for the of all-*trans*-β-carotene in the *Acrocomia aculeata* oil was verified through mathematical modelling. On the one hand, the use of a kinetic mechanism with more than one reaction step is unnecessary since it results in an overparameterized model. On the other hand, the model developed with just one reaction channel can simulate all the experimental conditions directly and effectively, it adequately fitted the available data within the experimental errors. The activation energy and the pre-exponential factor calculated from the Arrhenius clearly shows the dependence of the reaction rate constant k_{A*B} on temperature. The developed model with a unique reaction can be suggested to be used as an effective tool to optimise process conditions not only in laboratory but also on industrial scale. This study has provided further information enabling better comprehension of the kinetics of thermal degradation of macauba oil, which is a promising source of high-quality raw materials.

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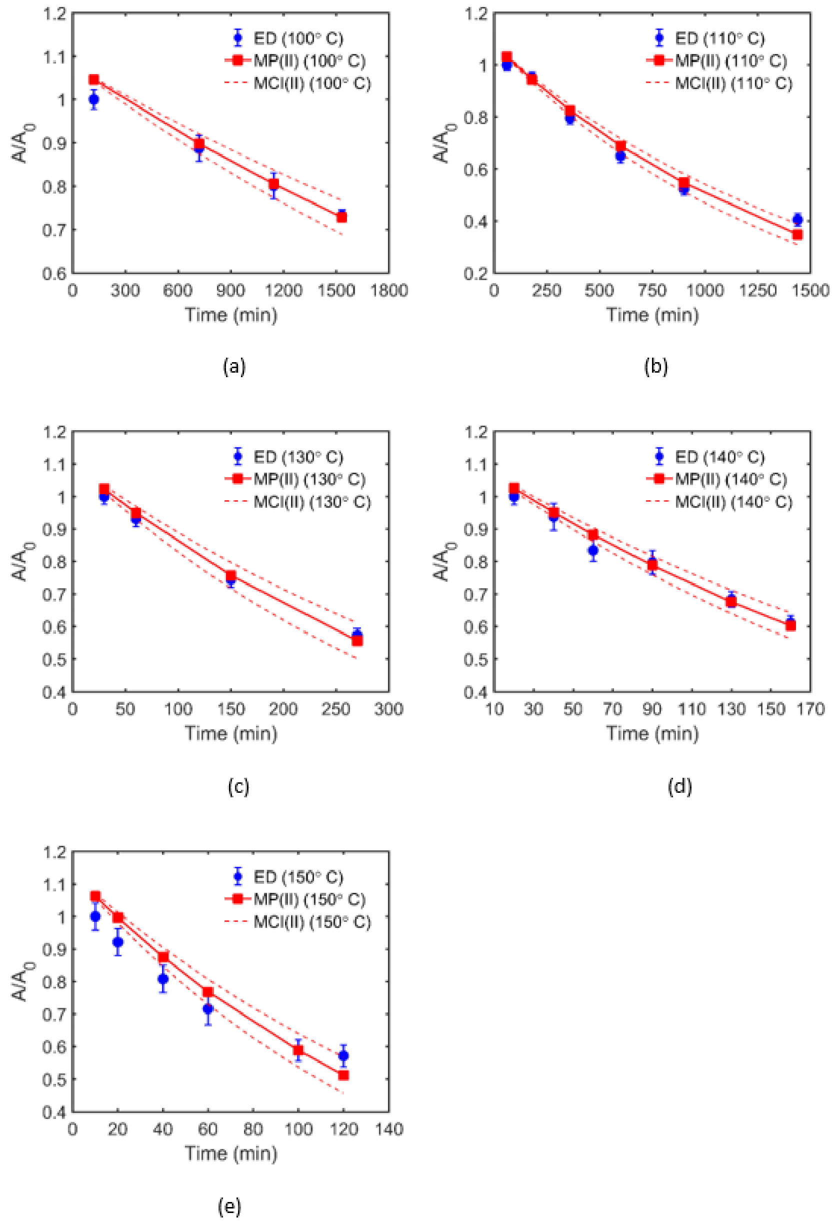

Figure 4: Predicted and experimental data at (a) 100, (b) 110, (c) 130, (d) 140 and (e) 150 ^{o}C using model derived from Mechanism II (MCI(II) = model confidence interval using Mechanism II; MP(II) = model predictions using Mechanism II; ED = experimental data).

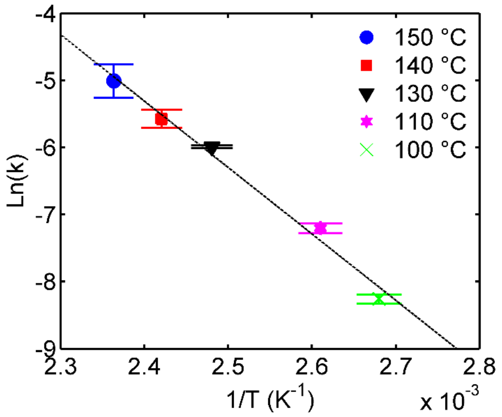

Figure 5: Arrhenius plot for the estimated model parameters at different thermal degradation temperatures using Mechanism II.

carry out this research.

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Continuous Stirred Tank Reactor: A Process Design for Interesterification of Macauba (*Acrocomia aculeata*) palm oil

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Abstract

Other than the edible oils extracted from the *Acrocomia aculeata* fruit, there is a growing interest in the palm to generate other high value-added products. Relatively high amounts of carotenoids (up to 378 mg kg^{-1}) have been found in the esculent oils mechanically obtained from the fruit mesocarp. From industrial application perspectives, several processes have been proposed to modify native vegetable oils to yield high functional properties of structured lipids. For interesterified products, the thermal effects of industrial reactors are crucial in reaction mechanisms. The present study has taken into account previously estimated kinetic parameters for the overall disappearances of all-trans β-carotene in the *Acrocomia aculeata* oil (k_o= 2.6 x 10^{-4} min^{-1}; Ea = 105.0003 kJ mol^{-1}; ΔH = 9.8 x 104 J kg^{-1}) to develop a continuous stirred tank reactor (CSTR) kinetic treatment that obeys first-order kinetics. A system of ordinary differential equations – mass and energy balances – was solved by the 4th order Runge-Kutta method (GNU Octave software). Under research conditions related to interesterification processing (2 h; 393.15 K), the initial concentration of carotenoids (around 11%) showed no significant decrease. Overall, realistic processing effects and conditions have been assessed, integrating results and knowledge, improving prospects of *Acrocomia aculeata* as a promising source of high-quality raw material, for producing functional ingredients and food with nutraceutical properties.

Keywords: Mathematical Modelling; CSTR; Runge-Kutta; Kinetics; Carotenoids; Interesterification

1 Introduction

Several industrial processes have been proposed to modify crude vegetable oils to meet the demands of contemporary consumers. These modifications are usually carried out to address functional characteristics of structured lipids (Xie & Chen, 2014; Xie, Yang & Zang, 2015; Xu, 2000). Among the most commonly applied methods to tailor physicochemical properties of edible oils, interesterification has received considerable attention. Unlike hydrogenation, interesterification processes are not related to the formation of trans fatty acids, therefore, extending the commercial application of modified lipids, including those with functional properties (Zhang, Lee, Zhou & Wang, 2019).

Interesterification processes can be carried out

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chemically or enzymatically, to reposition fatty acids on triacylglycerol structures. The process consists of ester breakages to form new aleatory bonds. Chemical interesterification is a process in which low-acid vegetable oils are not necessarily required to be previously bleached. Being relatively inexpensive when compared to enzymatic interesterification, the method is amenable to be scaled up being both efficient and feasible (Dijkstra, 2015; Tourchi Rudsari, Najafian & Shahidi, 2019).

Increasing demand by consumers and industries for new sources of natural food and products has been shown. The consumption of these products tends to be associated with health-related benefits obtained from bioactive compounds, among which fatty acids, carotenoids, and micronutrients constitute essential classes (Babbar, Oberoi & Sandhu, 2015; Cataldo, López, Cárcamo & Agosin, 2016; Oloo, Shitandi, Mahungu, Malinga & Ogata, 2014).

With a similar productive potential to *Elaeis guineensis*, Macauba (*Acrocomia aculeata*) is a high oil-yielding plant suited to edaphoclimatic zones, conditions adverse to African palms. An adult palm fructifies continuously with productivity from 4 to 6 tonnes of esculent oil per hectare. The oil extracted from the mesocarp is edible, containing up to 378 mg kg^{-1} of total carotenoids, with no antinutritional factors. It has a predominance of unsaturated fatty acids (77%), of which 53% and 18% are oleic (ω-9) and linoleic (ω-6), respectively. Contrarily, the oil extracted from the kernel has a predominance of saturated fatty acids (74%), of which 44% and 9% are lauric and palmitic, respectively (Evaristo et al., 2016; Nunes, Favaro, Galvani & Miranda, 2015).

From the perspective of lipid processing and structuring lipids, Macauba oils can be used as raw materials for producing different blends of vegetable oils to enhance the nutritional characteristics of structured lipids with specific functional properties. In this respect, thermal treatment control is essential for the retention of several bioactive compounds in vegetable oils (Pardauil et al., 2017; Tourchi Rudsari et al., 2019; Zhang et al., 2019).

The rate of degradation reactions tends to increase dramatically during thermal treatment mainly due to heating time and variations in temperature upsurges, particularly for carotenoids. Oxidation and cleavage products, in addition to 9-cis-β-carotene and 13-cis-β-carotene isomers, are major compound alterations that occur during thermal processing (Achir, Randrianatoandro, Bohuon, Laffargue & Avallone, 2010; Knockaert et al., 2012; Sampaio et al., 2013).

Previous kinetic evaluation of carotenoids degradation in vegetable oil importantly indicates a first-order reaction kinetic mechanism (Aparicio-Ruiz, Isabel Minguez-Mosquera & Gandul-Rojas, 2011; Knockaert et al., 2012). Through mathematical and predictive modelling and simulation, industries can benefit from the use of kinetic data and procedures to enhance knowledge and understanding of thermal food processing (Dinh, Sun & McLean, 2016; Singh, Singh & Ramaswamy, 2015).

The present study was undertaken primarily to develop a continuous version of the kinetic treatment of total carotenoids degradation reactions, through mathematical and predictive modelling, considering the thermal effects of conventional processes conditions usually applied for the interesterification of vegetable oils.

2 Materials and Methods

2.1 Crude Oil: *Acrocomia aculeata*

Acrocomia aculeata fruit was collected from native palms within a maximum of five days after the fall in the area of the Federal University of Minas Gerais - UFMG, in the metropolitan region of Belo Horizonte, Minas Gerais, Brazil. The mesocarp and kernel portions were promptly separated from the fruit, immediately frozen to a temperature of -18 ^{o}C and stored for up to 40 days. The mesocarp and kernel portions were thawed at room temperature, air-dried at 60 ^{o}C for 48 hours, and comminuted in a stainless-steel industrial blender (Omcan 31502, Mississauga, Canada) (Goula, 2013; Valério, 2017). The mesocarp and kernel pressings were performed on different days, to avoid cross-contamination.

Crude edible oils were then mechanically obtained from the mesocarp and kernel of *Acrocomia aculeata* fruit by continuously operated Expeller® press coupled to an electric motor (Sew-Eurodrive AAF67 DZ100LS4, Mealhada, Portugal). Amber glass vials (15 mL) were filled to the brim with the samples, minimizing the impact of light and oxygen intrusion by reducing the volume of headspace. Samples were stored at freezing temperature (-18 ^oC) until the analysis, to minimize possible rates and extents of enzymatic lipolysis, also concerning potential losses of antioxidants (Koidis & Boskou, 2015; Parducci & Fennema, 1978). For setup purposes, the content of total carotenoids in the crude *Acrocomia aculeata* mesocarp oil was defined as 378 mg kg^{-1}. The oil specific heat and density were stated as 107400 (J kg^{-1} min^{-1}) and 900 kg m^{-3}, respectively (CETEC, 1983; Fasina & Colley, 2008; Nunes et al., 2015).

2.2 Fatty acid compositions

Fatty acid compositions were determined based on the methods previously optimized by Christie (1989) and Guo, Hu, Qian and Wu (2012). The analysis was carried out on a GC 2010 System (Shimadzu, Japan) fitted with a Flame Ionisation Detector. Quantification of individual fatty acids methyl esters – FAME was conducted with a standard mixture of 37 esters of fatty acids (Supelco, Bellefonte, Pa., USA). The analysis was performed in triplicate. Samples were compared by Tukey multi comparison test ($p < 0.05$). Results were expressed considering the mean and sample standard deviation for the fatty acid composition, being expressed as a percentage of total fatty acids. The positive square root of variance formula is given according to the following Equation 1.

$$S = \sqrt{\frac{\sum(X_i - \tilde{X})^2}{n-1}} \tag{1}$$

In the above equation, S is the sample standard deviation, X_i is the sample measurement, $\tilde{X}$ is the sample arithmetic mean and n is the number of replicates.

2.3 Acid value

The Acid Value (AV) for the *Acrocomia aculeata* mesocarp oil was determined according to the AOCS Official Method 3d Cd-63 (AOCS, 2009). Determinations were performed by diluting 1 g of oil in 50 mL solution of isopropanol:toluene (1:1) followed by titration with potassium hydroxide (0.1 mol L^{-1}) standardized with potassium biphthalate. Values were calculated by the Equations (2) and (3) and presented; considering the mean and standard deviation, for the analysis performed in triplicate.

$$AV\ (mg\ KOH.g^{-1}) = \frac{(A-B) \times M \times 56.1}{W} \tag{2}$$

$$AV\ (\%\ oleic\ acid) = \frac{(A-B) \times M \times 28.2}{W} \tag{3}$$

In the above equations, A and B are the volume (mL) of potassium hydroxide (KOH) used for the sample and blank titration, respectively. M is the molarity of the applied basic solution, after standardization, and W is the sample weight (g).

2.4 Apparent kinetic and thermodynamic parameters

Apparent kinetic and thermodynamic parameters were previously estimated in a batch system (Valério, 2017), regarding the overall disappearances of total carotenoids naturally present in the mesocarp oil. The activation energy (E_a) of 105000.3 J mol^{-1} and the rate constant (k) of 2.6×10^{-4} min^{-1} were estimated by nonlinear regression, also considering the Arrhenius law. The enthalpy of activation (ΔH) of 9.81 x 104 J kg^{-1} was obtained by linear regression, according to the activation complex theory (Uzun & Ibanoglu, 2018). The parameters were then applied to the continuous version of the kinetic treatment, as seen in Table 1 of the following item 2.5.

2.5 Mathematical and predictive modelling: mass and energy balances

The mathematical and predictive modelling applied to the continuous version of the kinetic

Table 1: Process parameters: simulation data for CSTR

Parameter	**Parameter Identification**	**Unit**
F	Mass Flow	0.001 kg min^{-1}
C_{ao}	Carotenoids concentration (inlet)	378 mg kg^{-1}
R	Universal Molar Gas Constant	10.06 J kg^{-1} K^{-1}
To	Temperature of the Feed	393.15
V^a	Reactor Volume	100 dm^3
ρ	Oil Density	900 kg m^{-3}
cp	specific heat	107400 J kg^{-1} min^{-1}
q' = dq/dt	Flux Heat	1 x 10^3 kJ min^{-1}
E_a	Activation Energy	105000.3 J mol^{-1}
k_o	Rate constant	2.6 x 10^{-4} min^{-1}
ΔH	Enthalpy of Reaction	9.81 x 104 J kg^{-1}

[a] Volume was arbitrarily defined as 100 dm^3 for design purpose

modelling was performed through experimental and computer simulation techniques, using a numerical solution for a system of differential equations. The overall disappearance of all carotenoids forms in the mesocarp oil was studied to determine the intensity of the tetraterpenoids oxidative reactions in a Continuous Stirred Tank Reactor – CSTR (Achir et al., 2010; Sampaio et al., 2013). Mass and energy balances were conducted according to the following Equations 4 and 5, respectively, obeying the first-order formalism (Aparicio-Ruiz et al., 2011; Knockaert et al., 2012).

$$FCa_o - FCa - k_0 e^{\frac{-Ea}{R(Ta-T0)}} CaV = V\frac{dCa}{dt} \quad (4)$$

$$\rho FCpTo - \rho FCpTa - \Delta H k_0 e^{\frac{-Ea}{R(Ta-T0)}} V + q' = \rho VCp\frac{dTa}{dt} \quad (5)$$

In the above equation, F is the mass flow rate, C_{ao} and C_a are the total carotenoids concentration at the reactor inlet and outlet. k_o is the rate constant at the reference temperature, E_a is the activation energy, R is the universal molar gas constant. T_o is the temperature of the feed to the CSTR, T_a is the temperature of the underflow of the CSTR, V is the reactor volume, ρ is the oil density, Cp is the specific heat, ΔH is the enthalpy of reaction, and q' is the flux heat.
The equations presented were solved by a 4^{th} order Runge-Kutta method (GNU Octave Software, version 4.2.2 2018), from the perspective of developing and sharing both a free language program and a simple and effective solution algorithm. Thermal processing parameters were defined based on Grimaldi, Gonçalves and Ando (2005), Norizzah, Chong, Cheow and Zaliha (2004), Rodríguez, Castro, Salinas, López and Miranda (2001) and Petrauskaite, De Greyt, Kellens and Huyghebaert (1998). The isothermal temperature of 393.15 K was considered for the proposed time-dependent profile Dinh et al. (2016). The parameters for the CSTR simulation are presented in the following Table 1.

3 Results and Discussion

3.1 Fatty acid composition

Table 2 shows the mean and the standard deviation for the fatty acid composition (expressed as a percentage of total fatty acids) for the oils extracted from the *Acrocomia aculeata* mesocarp and kernel.
As it can be observed, **oleic** acid is predominant in the mesocarp oil. The level is similar to those (48-74%) of palm stearin (CAC, 2015), which is widely used for interesterification purposes. The **palmitic** acid was the second most abundant fatty acid in the oil extracted from the fruit mesocarp, which is in agreement with previous reports (Hiane, Ramos Filho, Ramos & Macedo, 2005; Nunes et al., 2015). Lauric acid was the predominant fatty acid in the Macauba kernel, as in

```
function f=continuosreactor(t,Y)
F=0.001; \% L.min-1
V=100; \% L
Ca0=378; \% (mg.kg-1)
k0=2.6e-4; \% (min-1)
dens=900; \% (kg.m-3 ou g.L-1)
cp=107400; \% J.kg-1.min-1
dif=((1/Y(2))-(1/393));
Ea=105000.32; \% J.kg-1
R= 10.06; \% J.kg-1.K-1
Deltah=9.8053e4; \% J.kg-1
if t$<$1.7
qadd=1e6 \% 103 kJ.min-1
else
qadd=0;
end
k=k0*exp(-(Ea/R)*dif);
f(1,1)=((F/V)*(Ca0-Y(1)))-k*Y(1);
aux=dens*V*cp;
f(2,1)=dens*F*cp*(373-Y(2))/aux+((Deltah*k*Y(1)*V)+qadd)/aux;
```

Figure 1: Algorithm: insertion of simulation data and equations

```
[tempo,solucao]=ode45('continousreactor',[0 5],[378,393]);
figure conc=solucao(:,1);
plot(tempo,conc,"Color","b","linewidth",4);
grid on;
set (gca, "fontsize",16);
xlabel("Time (minutos)", "Color","k","fontsize", 14);
ylabel("Carotenoids concentration (mg/kg)", "Color","k","fontsize",14);
figure temp=solucao(:,2);
plot(tempo,temp,"Color","r","linewidth",4);
grid on;
set (gca, "fontsize",16)
xlabel("Time (minutos)", "Color","k","fontsize", 14);
```

Figure 2: Computational program: Runge-Kutta solution.

other palm crops such as *Attalea speciosa* and in the kernel oil, kernel olein and kernel stearin of *Elaeis guineensis* (CAC, 2015).

The predominance of unsaturated fatty acids (MUFA: 62.11 ± 0.84%; PUFA: 11.37 ± 1.23%) was observed in the mesocarp oil. On the other hand, the predominance of saturated fatty acids (SFA) became apparent in the kernel oil (63.75 ± 0.84%). Importantly, the levels of SFA, MUFA and PUFA were significantly different, according to Tukey's test ($p < 0.05$), when comparing the mesocarp and kernel oils. Regarding the ω-6 family of fatty acids, the linoleic acid in the Macauba mesocarp (8.81%) and kernel (3.94%) were similar to those reported (CAC, 2015) for palm oil (9.0-10.0%), palm stearin (3.0-10.0%), sunflower oil (2.1-17.0%) and virgin and refined olive oils (3.5-21.0%). The contents of linoleic acid in the *Acrocomia aculeata* oils were higher than those registered for palm kernel oil (1.0-3.5%), palm kernel olein (2.4-4.3%) and palm kernel stearin (0.5-1.5%).

The clear distinction between the fatty acid profiles of *Acrocomia aculeata* mesocarp and kernel oils, especially regarding the contents of saturated and unsaturated fatty acid, already renders it an essential role as a potential alternative oil crop for interesterification purposes. The fatty acid compositions of these two types of oils are similar to other raw materials commonly blended to meet and improve functional performances of structured lipids (Xie & Chen, 2014; Xie et al.,

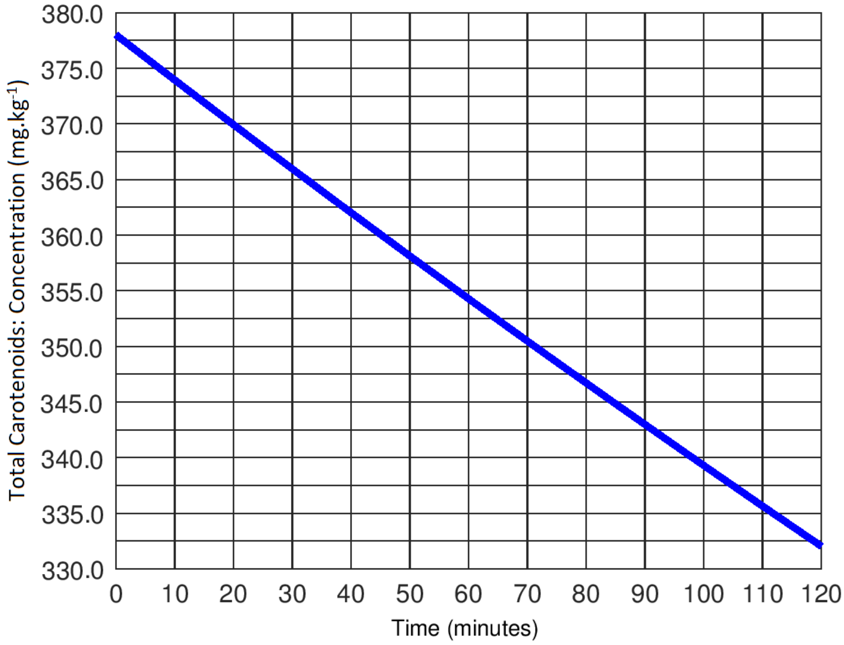

Figure 3: Runge-Kutta Solution: concentration profile for total carotenoids (Range time: 0 to 120 min)

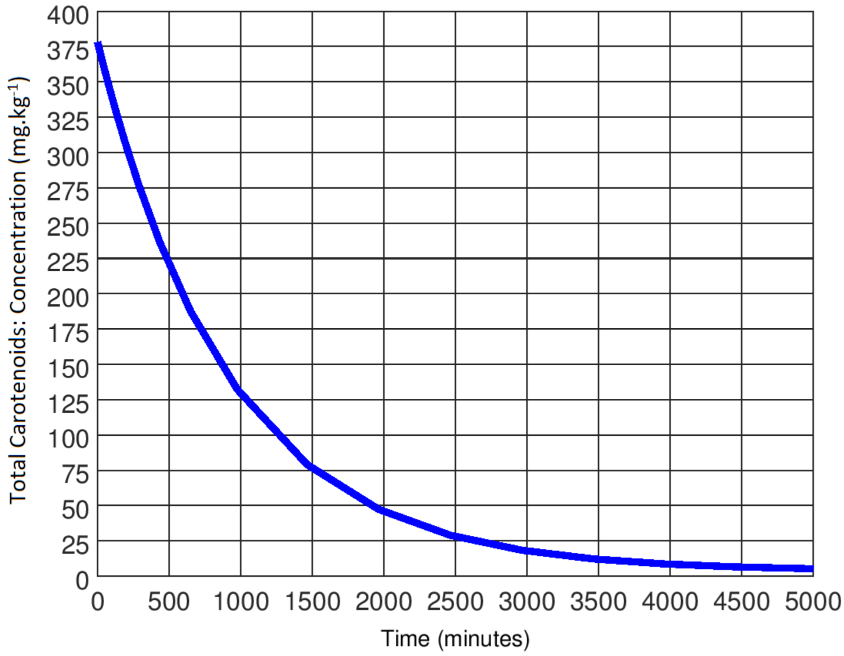

Figure 4: Runge-Kutta Solution: concentration profile for total carotenoids (Range time: 0 to 5000 min)

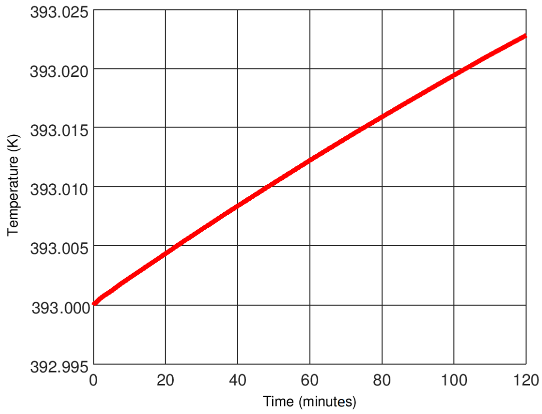

Figure 5: Runge-Kutta Solution: temperature profile for CSTR (Range time: 0 to 120 min)

2015; Xu, 2000; Zhang et al., 2019).

3.2 Acid value

Table 3 shows the mean and standard deviation for the acid values in the mesocarp oil.
As can be observed, the contents of free fatty acids (FFA) were well below the regulatory limits of up to 4 mg KOH.g^{-1} for virgin oils for human consumption (CAC, 2015; MAPA, 2006). Also, the acid value (0.8%, oleic acid) was below the acceptable limit of 3% economically applied for palm oils. It should be noted that the acid value has already been widely determined in literature for the Macauba kernel oil. It is, therefore, notable that the FFA in kernel oils range from 0.3 to 0.7 (mg KOH.g^{-1}), which are usually lower than that of mesocarp oils. In fact, lipolytic enzymes catalyze the decomposition of triglycerides which tends to be most active in mesocarps (CETEC, 1983; Coimbra & Jorge, 2011; Hiane et al., 2005). Low values of acidity observed for the oils extracted from the Macauba mesocarp and kernel adheres to interesterification purposes, raising the prospects of utilizing *Acrocomia aculeata* as an alternative source of high-quality raw material, for multi-purpose employments (Nunes et al., 2015). The low acid value observed also enhances chemical interesterification as an appropriate process for the crop productive chain, once it is considered amenable to be scaled up, efficient, and feasible to the edible oil industry in general (Tourchi Rudsari et al., 2019; Zhang et al., 2019).

3.3 Mathematical and predictive modelling

Figure 1 presents the algorithm developed to run the simulation data supported by the energy and mass balance equations in the GNU Octave Software. Figure 2 describes the algorithm that solves the presented system of differential equations, by 4^{th} order Runge-Kutta method. It is worth noting that that the Runge-Kutta method has shown stability properties, which has been effective and practical for solving the equations that describe the continuous system, which presenting convergence for the purposes of the present study.
From the perspective of developing a free language program, it should be reasonable to suggest that Figure 1 and Figure 2 might also allow expanding aspects of kinetic applications by sharing solution that complements batch estimations. It is noteworthy that the Octave Program software is compatible with conventional computer systems. It is therefore suggested that its use may not be only a practical solution but also a future possibility for innovative research and development.
Simulation models have been increasingly used to solve problems and to aid in decision-making. Results obtained from computational simulation tend to contribute with practical advantages that also consider predictive capabilities for industrial projects and processes, and to possibly combine raises of efficiency, productivity, and cost reduction.
The total carotenoids concentration and temperature profiles for CSTR are represented by Figures 3, 4 and 5, resulting from simulations.
Figure 3 shows that the total carotenoids concentration profile was continually decreasing regarding the CSTR inlet, starting from C_{ao} equal to 378 mg kg^{-1}. The temperature profile, shown in Figure 5, was ascending by the addition of flux heat (10^3 kJ min^{-1}). As can be observed, the addition of 1000 kJ to the process slightly increases the accumulated temperature. Thermodynamic

Table 2: Fatty acid compositions expressed as a percentage of total fatty acids

Fatty Acid	*Acrocomia aculeata*	
	Mesocarp Oil	**Kernel Oil**
Caproic acid (C6:0)	ND*	0.35 ± 0.00^{i}
Caprylic acid (C8:0)	0.08 ± 0.05^{g}	4.38 ± 0.04^{e}
Capric acid (C10:0)	ND*	3.62 ± 0.03^{g}
Undecylic acid (C11:0)	0.08 ± 0.04^{g}	ND*
Lauric acid (C12:0)	0.04 ± 0.01^{g}	37.22 ± 0.03^{a}
Myristic acid (C14:0)	0.07 ± 0.00^{g}	8.12 ± 0.02^{c}
Pentadecylic acid (C15:0)	0.17 ± 0.00^{g}	0.03 ± 0.00l
Palmitic acid (C16:0)	19.62 ± 0.56^{b}	6.88 ± 0.02^{d}
Palmitoleic acid (C16:1)	1.68 ± 0.04^{ef}	0.16 ± 0.01^{j}
Margaric acid (C17:0)	0.09 ± 0.00^{g}	0.04 ± 0.00l
Ginkgolic acid (C17:1)	0.06 ± 0.00^{g}	0.04 ± 0.00l
Stearic acid (C18:0)	5.15 ± 0.17^{d}	2.90 ± 0.02^{h}
Oleic acid (C18:1)	60.33 ± 1.18^{a}	32.03 ± 0.01^{b}
Linoleic acid (C18:2)	8.81 ± 0.32^{c}	3.94 ± 0.02^{f}
α-Linolenic acid (C18:3)	0.73 ± 0.02^{fg}	0.06 ± 0.00^{kl}
Arachidic acid (C20:0)	0.21 ± 0.00^{g}	0.14 ± 0.00^{jk}
Gadoleic acid (C20:1)	0.07 ± 0.01^{g}	0.14 ± 0.01^{jk}
Behenic acid (C22:0)	0.14 ± 0.01^{g}	0.09 ± 0.01^{jkl}
Eicosadienoic acid (C20:2)	1.83 ± 0.46^{ef}	ND*
$\sum SFA^{**}$	25.65 ± 0.84	63.75 ± 0.06
$\sum MUFA^{***}$	62.14 ± 1.23	32.32 ± 0.01
$\sum PUFA^{****}$	11.37 ± 0.80	3.93 ± 0.01

The results represent the mean ± standard deviation of the analysis performed in triplicate.
Values within the same column – means, followed by the same letter, do not differ by Tukey's test ($p < 0.05$).
*ND: Not Detected
Note: **) Saturated Fatty Acids; ***) Monounsaturated Fatty Acids; ****) Polyunsaturated Fatty Acids.

Table 3: Acid value: *Acrocomia aculeata* mesocarp oil

Process Parameters	**Dimensions**
Acid Value (mg KOH g^{-1})	1.6 ± 0.1
Acid Value (% oleic acid)	0.8 ± 0.1

variations were not numerically significant at 120 minutes, due to the endothermic nature of the reaction (Knockaert et al., 2012). For comprehensiveness purposes, Figure 4 shows the thermal degradation profile of total carotenoids in the *Acrocomia aculeata* oil, over 5000 minutes of thermal processing. The half-life time reaction ($t_{1/2}$) can be observed at 750 minutes of processing.

From Figure 3 it can be observed that the total carotenoids concentration in the *Acrocomia aculeata* mesocarp oil varied from around 6.7% at the first 60 min ($Ca_{(60min)}$: 354.6 mg kg^{-1}), ranging to a maximum degradation of 12.6 % ($Ca_{(120min)}$: 331.8 mg kg^{-1}) at the end of the proposed processing. It should be noticed that the research conditions (40 minutes; 393.15 K) similar to those of interesterification decrease the initial amount of total carotenoids (4.22 %) in a CSTR.

Carotenoids losses during food processing have been reported and quantified by previous studies (Mader, 1964; Magosso et al., 2016; Onyewu, Ho & Daun, 1986; Palmero et al., 2013), although, not often encompassing the effects of processing factors on the thermal degradation. Comparisons between kinetic predictions may become significant when the parameters estimated by batch studies are added to the context. When considering kinetic parameters given by previous studies (Achir et al., 2010; Knockaert et al., 2012), a reduction by around 70% and 80% in the carotenoid concentration found for batch systems. Yet, after 60 minutes around 57% (215 mg kg^{-1}) of the total carotenoid would be degraded as a consequence of temperature supply.

Findings suggest that carotenoid degradation is complex and highly dependent on factors linked to the systems in which they are contained. Hence, process optimization does require product-specific kinetic data. It is essential to consider that process optimization must be supplemented with thermodynamic information for designing efficient processes at industrial scale reactors (Colle, Lemmens, Van Buggenhout, Van Loey & Hendrickx, 2013; de Carvalho et al., 2020). The simultaneous mass and energy balance, to some extent, brings a significant approach to the real application as proposed in this study.

The highly unsaturated structures of carotenoids make the compounds considerably sensitive to thermal degradation reactions. The rates of degradation tend to increase during thermal treatments as temperature rises. Once the typically used temperature in industrial chemical interesterification processes ranges from 60 to 120 ^{o}C, it is recommended to work at milder temperature ranges for extended periods, instead of higher temperatures, in this ambit, for a shorter, to obtain a processed *Acrocomia aculeata* oil with preserved natural carotenoid content (Achir, Penicaud, Avallone & Bohuon, 2011; Pardauil et al., 2017; Rodriguez-Amaya, Rodriguez & Amaya-Farfan, 2006).

The use of *Acrocomia aculeata* oils clearly agrees with efforts for producing different blends of vegetable oils to improve functional performances of structured lipids (Tourchi Rudsari et al., 2019; Xie & Chen, 2014; Xie et al., 2015), in reference to the increasing demand for natural food and products (Babbar et al., 2015; Cataldo et al., 2016; Oloo et al., 2014).

4 Conclusion

A continuous version of the kinetic treatment of total carotenoids degradation that follows a first-order model has been developed that takes into consideration the thermal effects of conventional processes conditions usually applied for the interesterification of vegetable oils. The findings demonstrate the success of applying the 4th order Runge-Kutta method to describe the interesterification of Macauba oil, in a CSTR, with minimum carotenoid degradation. Overall, trends of milder degradation tend to be observed when thermal processes are carried out in a continuous system, which has a relatively shorter residence time when compared to the same processes conducted in batch systems. By solving the mass and energy conservation balances, applying the Runge-Kutta method, it was possible to observe the concentration and thermal profile curves. It was possible to verify that the total carotenoids concentration profile was decreasing. The accumulated temperature, in turn, increased slightly, due to the exothermic nature of the reaction. The initial concentration in the oil has not significantly decreased (4.22 %) when considering time-temperature conditions (40 min; 393,15 K) similar to those of interesterification processes. Evaluations of pilot plant installations should be considered for future studies to validate simulation results presently addressed. Experimental data concerning interesterification may be added for future approaches and procedures, again emphasizing the importance of numerical predictions for practical applications, to contribute to a more realistic industrial design.

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Consumers' Perception and Consumption of Sunflower Oil in Kumasi, Ghana

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Abstract

Consumption pattern among indigenous groups is usually influenced by consumers' sociocultural and perceptual factors. This study employs the binary logit model to analyse the factors that influence the consumption of sunflower oil in Kumasi, Ghana. A cross-sectional approach was used to obtain data from 200 consumers who were selected using a multi-stage sampling method. The results showed that a majority (93%) of the respondents were aware of the availability of sunflower oil on the local market and a third (69.5%) had used it for cooking before. The respondents agreed with the perception statements that sunflower oil is healthy, expensive, reduces the risk of heart diseases and cancer and has better frying performance. The empirical results of the logit regression model showed that consumption of sunflower oil is influenced by household size, awareness of the product and perceptions on health benefits, price and frying performance of sunflower oil. The price of the oil was identified as the most important constraint to its use albeit it had no negative effect on its consumption. Investments in the production and promotional strategies on the use of sunflower oil should consider the significant variables that have influence on its consumption.

Keywords: Sunflower oil; Perception; Consumption; Logit regression; Kumasi-Ghana

1 Introduction

Sunflower (*Helianthus annuuss* L.) is one of the few crop species that originated in North America (most originated in the fertile crescent of Asia, South or Central America) (Department of Agriculture, Forestry and Fisheries. (DAFF), 2010). Sunflower is a plant of the compositae family, a large family of flowering plants that are mainly herbs and shrubs. The plant's tolerance to drought and great variety of soils accounts for its suitability to most areas. The seeds of sunflower are rich in oil and it is considered to be a potential source of protein for human consumption; this due to its high ratio of polyunsaturated/saturated fatty acids and the high content of linoleic acid (Ohlson, 1992).

The oil extracted from sunflower seeds is a good source of oil for cooking and manufacturing of margarine, paints, soaps, and cosmetics as well as biodiesel. Oil concentration of sunflower seeds is about 44%, on average (Andrianasolo, Debaeke, Champolivier & Maury, 2016). In addition to the oil, edible proteins can be obtained from the cake for human consumption. The cake is also a good source of protein in the manufacture of furfural in yeast, alcohol production and as fuel. Sunflower oil production and distribution has been on the rise worldwide in recent years due to consumers becoming more particular about the food they eat and how it affects their health

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and lifestyles. The Ghanaian consumption patterns have changed toward western (European and American) foods as a result of urbanization, women working outside the home and a shift in the lifestyles of a large youth population. Middle-class incomes are also rising and there is now higher demand for healthy foods. A growing concern among Ghanaian consumers with regard to food safety and healthy diets is also increasing the demand for higher quality products. As a result, domestic processors are developing and improving food products in order to meet the needs of this niche market (Hammond, 2014).

Ideally, the preparation and consumption of healthy meals provides good health and proper growth of individuals. Edible oil is one of the most important components of food and a major source of energy for human survival. Most households in Ghana use edible oils in the preparation of their daily meals. They are particularly high in saturated fat which contributes to coronary diseases when consumed in excess (Prema, 2013). In past years, the general government health expenditure has increased from US$53 per capita in 1995 to US$60 per capita in 2014 (Adua, Frimpong, Li & Wang, 2017). Roberfroid (2000) reported that today's foods are intended not only to satisfy hunger and to provide necessary nutrients for humans but also to prevent nutritional diseases and improve physical and mental well-being of consumers. Sunflower oil is an excellent source of vitamin E (Warner, Vick, Kleingartner, Isaak & Doroff, 2003). Sunflower oil gives consumers an even healthier food supply and lifestyle as it meets the needs of consumers and food processors alike for a healthy and high performance non-transgenic vegetable oil. The vitamin E in sunflower oil is a required nutrient that functions as an antioxidant and decreases the risk of heart disease, cancer and bolsters immune function (Grigg, 1999).

However, in Ghana there is high consumption of other edible oils compared to sunflower oil due to factors which are not precisely known and also because sunflower oil is a relatively new product on the Ghanaian market. Generally, people have negative perceptions and usually dislike new products on the market because of the uncertainty of the attributes of those products and the uncertain utility to be derived from the consumption of such products. This may account for consumers' preference for already existing products that they are familiar with. Awareness about a product's health benefits is needed to enhance consumer decision making on available products. Consumers are inadequately informed about the perceived sunflower oil health benefits; this could be a hindrance to the consumption of sunflower oil. Consumers' perceptions of the attributes of a product play a vital role on their purchase and consumption decisions (Nimoh, Asare, Twumasi & Anaman, 2018). This study therefore sought to investigate consumers' awareness and perception of consumption of sunflower oil, and the factors affecting their consumption of the product as well the constraints to consumption in the Kumasi metropolis of Ghana. The study hypothesized that the consumption of sunflower oil is influenced by the socioeconomic characteristics of consumers such as age, education, household size and income, as well as their knowledge and perception about the product.

2 Materials and Methods

2.1 Conceptual framework

The study employed the random utility theory to explain a consumer's decision to consume sunflower oil among other edible oils available to the consumer. Random utility theory makes the assumption that every consumer is a rational decision-maker with the objective of maximizing his or her utility relative to the available choices and given constraints (Loureiro & Umberger, 2007). That is, the decision-maker i^{th} consumer in making a choice considers M_j mutually exclusive alternatives that constitute his or her choice set which may differ according to the decision-maker. The i^{th} consumer assigns to each alternative j in his choice set a perceived utility U_j^i and selects the alternative with the maximum utility (Anderson, De Palma & Thisse, 1992). The utility assigned to each alternative (choices) depends on a number of measurable characteristics or attributes of the said alternative and of the decision-maker, specified as:
$U_j^i = U_i^i\ (X_j^i)$, where X_j^i is the vector of attributes relative to alternative j and to the decision-

maker i. It is, however, acknowledged that consumers may take decisions that do not maximize their utility. This behaviour may result from errors in perception resulting from lack of information on product attributes or discounting inability or market failures such as price structure that do not reveal the real cost of the production to the society or limitation in the set of products available to the consumer (Tiffin, Bruce Traill & Mortimer, 2006).

2.2 Empirical model

The logit model has been used in studies to determine factors affecting consumers' willingness to consume a product. The logit model is preferred over other categorical variable estimation techniques (Maddala, 1986) and is a better procedure for capturing the magnitude of the independent variable effect for qualitative dependent variables than the probit models (Amemiya, 1985). The logit is estimated using maximum likelihood estimation as it results in large-sample properties of consistency and asymptotic normality of the parameter estimates.
The logit model, based on the cumulative logistic probability function, was used to analyze the effect of factors that influence the consumer's consumption of sunflower oil (Pindyck & Rubinfeld, 1988). The dependent variable is a dichotomous or binary choice, having two options of consuming sunflower and not consuming sunflower oil. The logit model is specified as follows:

$$Pi = F(Zi) = F(\alpha + \beta Xi) = \frac{e^{\alpha + bXi}}{1 + e^{\alpha + bXi}} \quad (1)$$

The influence of consumers' socioeconomic characteristics and perception attributes of sunflower oil on their consumption behaviour was analyzed with the simple logit regression model. From equation 1, the empirical specification of the logit model is given as:

$$\begin{aligned} Sunfloil_{ij} &= \beta_0 + \beta_1 \cdot gend_{ij} + \beta_2 \cdot age_{ij} + \\ &\beta_3 \cdot hdsize_{ij} + \beta_4 \cdot mincome_{ij} + \beta_5 \cdot education_{ij} + \\ &\beta_6 \cdot aware_{ij} + \beta_7 \cdot healthier_{ij} + \beta_8 \cdot expensive_{ij} + \\ &\beta_9 \cdot betterfrying_{ij} + \beta_{10} \cdot vitaminE_{ij} + \epsilon_i \end{aligned} \quad (2)$$

Table 1 presents the definition of explanatory variables used in the logit regression model.

2.3 Population, sampling and data collection

The target population was all edible oil consumers in Kumasi, Ghana. A multi-stage sampling technique was employed to select 200 consumers of edible oil from Kumasi metropolis. Kumasi metropolis was purposively selected because of its cosmopolitan nature. Three stage sampling was used to arrive at the respondents interviewed for the study. Stage one involved selection of sub-metros under Kumasi metropolis; stage two focused on selecting communities; and stage three focused on the respondents that participated in the survey. Four (4) sub-metros were purposively selected according to their income levels in the second stage. At the third stage, 50 vegetable oil consumers within each community were selected by using the systematic random sampling method based on every tenth residential address of the study communities. The face-to-face interview method was employed to obtain relevant responses from the sampled consumers using a semi-structured questionnaire made up of both open- and closed-ended questions. A pilot survey allowed for streamlining of the questions to achieve the objectives of the study. The face-to-face interviews provided the opportunity to explain the questions which were difficult for respondents to answer. This helped to obtain the accurate information required and also provided the opportunity for the interviewer to educate the respondents.

2.4 Analysis

Data were subject to both descriptive and inferential analyses. Data on the socio-economic characteristics of respondents and consumer evaluation of sunflower oil attributes were summarized with descriptive statistics, i.e. frequencies and percentages, graphs and mean scores. A perception index (PI) was computed based on a 5-point Likert scale (*1= strongly disagree to 5 = strongly agree*) to assess consumers' perception of consumption of sunflower oil. The logit regres-

Table 1: Definition of explanatory variables in the model

Variable	Description of variable	a priori sign
Dependent variable		
Sunfloil	Willingness to consume sunflower oil (Willingness to consume = 1, Otherwise = 0)	
Independent variables		
Gend	Gender *(Female =1, Male = 0)*	+
Age	Age of consumer *(Years)*	+/-
Hdsize	Household size *(Number of people in the household)*	+
Mincome	Consumer's monthly income *(GH¢)*	+
Education	Educational level *(Number years in formal education)*	+
Aware	Awareness of sunflower oil *(Yes =1, No = 0)*	+
Healthier	Perception that sunflower oil is healthier compared to other refined edible oils (Yes =1, No = 0)	+
Expensive	Perception that sunflower oil is expensive than the other refined edible oils (Yes =1, No = 0)	+
Better frying	Perception that sunflower oil has better frying performance than other oils (Yes =1, No = 0)	+
Vitamin E	Perception that sunflower oil contains Vitamin E that reduces the risk of cancer (Yes =1, No = 0)	+

sion model was used to analyse factors that influenced the consumption of sunflower oil. Constraints to the consumption of sunflower oil in the study area were analyzed using the ranking and the Kendall Coefficient of Concordance (W).

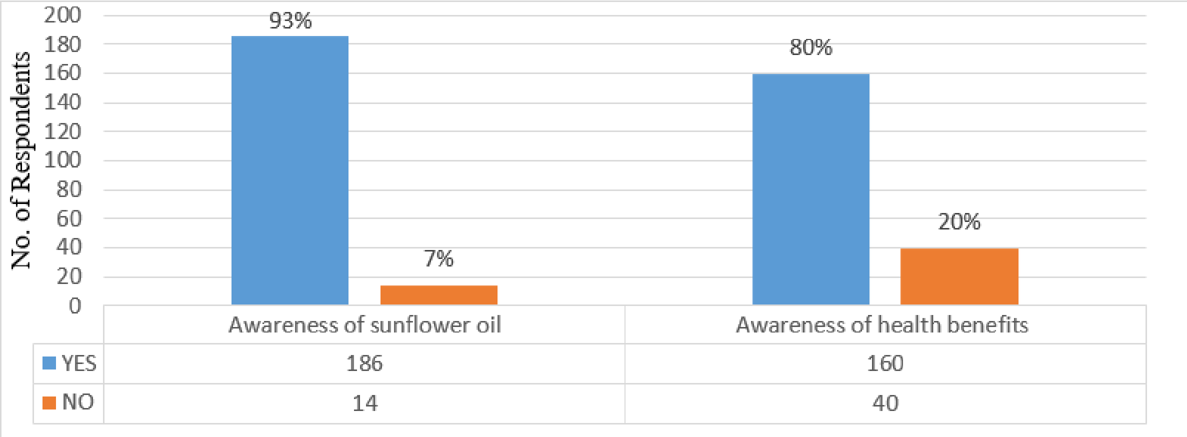
3 Results and Discussion

3.1 Socio-economic characteristics of respondents in the Kumasi Metropolis

The majority of the respondents (88%) were females (Table 2). This was due to the fact that there is high female participation in cooking in the traditional African household and that purchase of foodstuffs, including cooking oil is dominated by females in a household. The majority of the respondents (62%) were married. This finding agrees with Basorun (2009) that cooking remains the exclusive responsibility of women that are married. The majority (90%) of the respondents were Akans, the largest ethnic group in Ghana which comprise the Asante, Bono, Adanse, Twifo, Asen, Fante, Akuapem, Akyem, Akwamu, Kwahu, Sehwi, Awowin, Nzima and Ahanta. the majority of respondents were working as self-employed (71% of the sample). The mean age of respondents was 37 years, implying that most of the respondents were within the working age group. The size of a household to a large extent determines both the frequency and amount of food that the house can purchase. Normally, the larger the household size, the greater the amount of food consumed as compared to smaller ones. The mean household size was approximately five persons. According to Stewart, Harris and Guthrie (2004), household size and a large number of dependents partly influence food choices. The average number of years of education among the respondents was 11 years, representing secondary level education, which is higher than the average Ghanaian schooling years of 5.16 (Ghana Living Standards Survey, 2005), signifying that most of the consumers captured in this study had high level of education. The average monthly income of respondents was GH¢582.95.

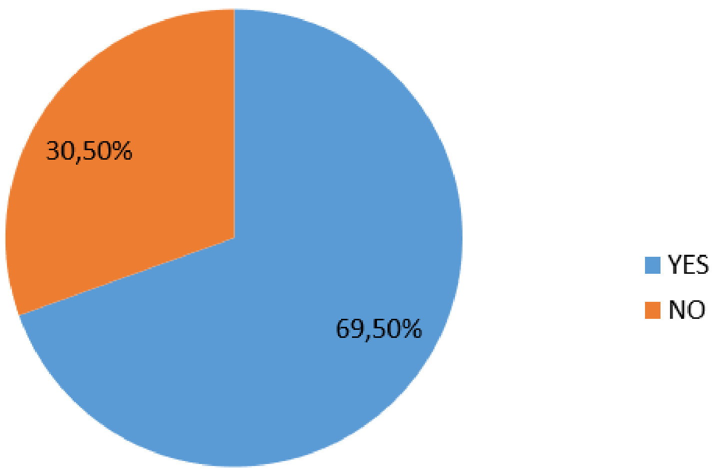
3.2 Consumers' awareness and patronage of sunflower oil

The majority of the respondents had knowledge about sunflower oil (93%) and about its health benefits (80%) (Figure 1), implying that consumers' awareness about sunflower could be an indication of a good market potential for the product. the majority of the respondents had purchased or used sunflower oil before (69.5%) (Figure 2), implying that most of the respondents were familiar with the product. The main reasons for the patronage of the product were



Source: Field survey, 2016

Figure 1: Awareness of sunflower and its health benefits



Source: Field survey, 2016

Figure 2: Patronage of sunflower oil

Table 2: Socioeconomic characteristics of respondents

Categorical Variables	**Description**	**Frequency**	**Percentage**
Gender	Male	25	12.5
	Female	175	87.5
Marital Status	Single	77	38.5
	Married	123	61.5
Ethnic Group	Akan	179	89.5
	Ewe	7	3.5
	Ga	2	1.0
	Northerners	12	6.0
Employment Type	Government	14	7.0
	Self Employed	142	71.0
	Private	16	8.0
	Student	15	7.5
	Unemployed	13	6.5
Continuous Variables	**Minimum**	**Maximum**	**Mean**
Age	19	84	37.35
Household Size	1	15	4.73
Years in formal education	0	19	10.49
Income	0	3000	582.95

Source: Field survey, 2016

mentioned as the health and nutritional attributes of the product. This finding agreed with that of Gracia and de Magistris (2008) for some consumers in south of Italy who indicated the health and nutrition perspectives as key in the purchasing of food.

3.3 Consumers' perception of sunflower oil

For consumers to accept and pay for a product, that particular product must possess attributes that are considered most important and attractive to them. Steenkamp and van Trijp (1996) suggested that a product's quality attributes form the basis for consumer preference for a product. The results from Table 3 showed that majority of the respondents agreed with the statements that sunflower oil is healthier compared to the other refined edible oil (70%, 3.83); is of higher quality than other refined edible oils (69%, 3.79); helps in reducing the risks of getting heart diseases (51%, 3.62); and has better frying performance than other oils (59.5%, 3.63). However, about one-third (29%, 3.32) of the respondents agreed with the statement that sunflower oil is rich in Vitamin E which reduces the risk of getting cancer, and majority (74.5%, 3.93) also agreed with the statement that sunflower oil is more expensive than other refined edible oils. Averaging the six scores gave an overall perception index (PI) of 3.69, implying a good perception of consumption of sunflower oil.

3.4 Factors affecting the consumption of sunflower oil

The study found a significant difference in some of the socioeconomic characteristics of the respondents who consume sunflower oil and those who do not consume, particularly with gender and awareness of sunflower oil as well as their perception of the product, except the perception that sunflower oil contains Vitamin E that can reduce the risk of cancer (Table 4). Based on the empirical results from the logit regression model (Table 5), the variables found to influence the consumption of sunflower oil were household size,

Table 3: Consumers' perception of sunflower oil

Statements	**Strongly disagree 1**	**Disagree 2**	**Undecided 3**	**Agree 4**	**Strongly Agree 5**	**Mean Score**
Sunflower oil is healthier than other refined edible oil	2	8	50	102	38	**3.83**
Sunflower oil is expensive than other refined edible oil	0	14	37	98	51	**3.93**
Sunflower oil is of higher quality than other refined edible oils	0	10	52	108	30	**3.79**
Sunflower oil helps in reducing the risks of getting heart diseases	0	2	96	78	24	**3.62**
Sunflower oil is rich in Vitamin E which reduces the risk of getting cancer	0	4	138	49	9	**3.32**
Sunflower oil has better frying performance than other oils	1	8	72	103	16	**3.63**
Perception Index						**3.69**

Source: Field survey, 2016

Table 4: Descriptive statistics of respondents in the Kumasi Metropolis

Independent variable	**Consume sunflower oil**	**Not consume sunflower oil**	**Mean difference**	**Significance**
Age	38.33	35.47	2.86	0.875
Gender	0.92	0.7941	0.12***	0.000
Household Size	4.86	4.49	0.37	0.595
Years in School	10.64	10.25	0.39	0.668
Monthly Income	597.80	400.44	197.36	0.768
Awareness	0.99	0.81	0.18***	0.000
Perception Statements				
Sunflower oil is healthier compared to other refined edible oils	0.86	0.40	0.47***	0.000
Sunflower oil is expensive than the other refined edible oils	0.88	0.50	0.38***	0.000
Sunflower oil has better frying performance than other oils	0.77	0.26	0.51***	0.000
Sunflower oil contains Vitamin E that reduces the risk of cancer	0.34	0.19	0.15	0.254

*, **, ***: significance at 10%, 5% and 1% respectively. Source: Authors' calculation, 2016

Table 5: Logit estimation of determinants of consumption of sunflower oil

Independent variables	**Coefficient**	**Standard error**	**dy/dx**	**P-value**
Age	0.0146	0.0180	0.0019	0.415
Gender	0.7909	0.5915	0.1046	0.181
Household size	0.1468*	0.0879	0.0194	0.095
Years in school	0.0560	0.0549	0.0074	0.308
Monthly Income	0.0003	0.0005	0.0000	0.573
Awareness	2.3728**	1.1919	0.3137	0.047
Sunflower oil healthier than other refined edible oils	1.6856***	0.4357	0.2229	0.000
Sunflower oil expensive than the other refined edible oils	1.0415**	0.4897	0.1377	0.033
Sunflower oil has better frying performance than other oils	1.1600**	0.4721	0.1534	0.014
Sunflower oil contains Vitamin E that reduces the risk of cancer	-0.1604	0.4639	-0.0212	0.730
Constant	-6.6893***	1.7866		0.000
Obs., 200; Pseudo R^2 , 0.3427; Log likelihood, -84.2692				

*, **, ***: significance at 10%, 5% and 1% respectively. Source: Authors' calculation, 2016

respondents' awareness of the product and all the perception variables considered, except the perception that sunflower oil contains Vitamin E that can reduce the risk of cancer.
The variable household size, had a positive and statistically significant effect at 10% for the consumption of sunflower oil. An increase in household size tends to increase the consumption of sunflower oil by 1.94%. This result is consistent with the finding by Ali, Aslam and Rasool (2013) who observed that household size significantly affects consumption edible oil. Also, awareness of sunflower oil as an edible oil product on the Ghanaian market was found to be positive and significant in affecting the consumption of sunflower oil at 5% level. This implies that a unit change in the awareness of sunflower oil will increase the tendency of consumption of the product by 31.37%. This finding agrees with result by Kathuria and Jit (2009) that the brand or product awareness is the consumer's ability to identify the brand and can be measured with the help of brand recall and brand recognition. In addition, consumers' perception that the product is healthier than other vegetable oils was positive and significant at 1%, implying that the positive perception about the product tends to influence its consumption by 22.29%. This supports the finding by Roberfroid (2000) who indicated that today's foods are not intended to only satisfy hunger and to provide necessary nutrients for humans but also to prevent nutrition-related diseases and to improve physical and mental wellbeing of the consumer. Therefore, consumers of sunflower oil view the product in respect of the disease preventing attribute as an important factor in deciding whether or not to patronize it. Moreover, the perception that sunflower oil has a better frying performance had a positive effect and tends to influence consumption of the product by 15.34%. This implied that the use/cooking property of sunflower oil is of great concern to consumers in their choice of edible oil. This finding was in agreement with the findings of Steenkamp (1997). In spite of the perception that sunflower oil is more expensive than the other refined edible oils, the variable had a positive and significant influence on its consumption at 10% significance level. A unit change in the perception that sunflower oil is expensive will influence the consumption of the product by 13.77%, implying the health consciousness of consumers irrespective of the price of the product.

3.5 Constraints to the consumption of sunflower oil

Table 6 presents results on the constraints to the consumption of sunflower oil. The majority of the respondents ranked price of sunflower oil as the most important constraint when pur-

Table 6: Constraints to the consumption of sunflower oil

Constraints	Most Important 1	More Important 2	Moderate Moderate 3	Slightly Important 4	Least Important 5	Mean Mean	Rank Rank
Price	124	22	11	1	2	1.43	1st
Advertisement	18	29	26	7	3	2.86	2nd
Preference	26	26	22	18	15	3.66	3rd
Package	5	32	22	25	18	3.72	4th
Availability	50	34	26	30	14	4.15	5th
Taste	4	27	10	9	23	5.17	6th
Kendall's W^a, 0.457; Chi-square, 139.237; Df., 5.00; Asymp. Sig, 0.000							

Source: Authors calculation, 2016

chasing sunflower oil, albeit this variable was not found to negatively affect the consumption of the product as per the empirical regression result (Table 5). Lack of advertisement or promotion of sunflower oil was found to be the second ranked concern to the low consumption of the product. The Kendall‘s Coefficient of Concordance (W^a) for the rankings of constraints faced by respondents was estimated at 0.457, implying agreement of about half of the respondents with the order of ranking of the constraints.

4 Conclusion

From the findings of the study, it can be concluded that most consumers in the study area were aware of sunflower oil and its health benefits, and thus willing to consume the product albeit with the perception that it is more expensive than the other refined edible oils. The logit regression model showed that consumption of sunflower oil was influenced by household size, awareness of the product, perceptions on health benefits, price and frying performance. Price was considered to be the important constraint to the consumption of sunflower oil although it did not have negative effect on its consumption. There is a need to promote the consumption of sunflower to help improve the health of consumers. However, producers need to consider the socioeconomic and perception factors influencing its consumption in an attempt to promoting its use. Manufacturers and retailers must promote and develop new product design and marketing strategies of the sunflower oil product based on its health benefits and frying performance whilst considering the price to increase their market share.

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Influence of Raw Meat Content on 3D-Printing and Rheological Properties

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Abstract

The aim of this study was to investigate the influence of raw chicken meat content on the rheological properties and 3D printability of minced meat mixtures using different concentrations of raw and cooked chicken meat. The meat mass contained yolk, crushed ice, lean raw meat and cooked meat with a high concentration of connective tissue. The concentrations of raw meat added to cooked meat as a percentage of the total weight of meat were 0; 30; 40; 50; 60; 70 and 100. To determine the rheological properties, amplitude sweep and frequency sweep were carried out with a Rheostress RS 300 (Thermo Fisher Scientific Inc.). Cubes were printed, and the printability and optical impression were evaluated using grades from 1-5. The results showed that rheological properties had a strong influence on the printability of meat mass and it is necessary for G' (storage modulus) at the LVR (linear viscoelastic region) to be higher than 7000 Pa. The complex viscosity $\mid \eta^* \mid$ should be higher than 170 Pa, at a shear stress $\tau = 10$ Pa, and a frequency f = 10 Hz used to guarantee sufficient solidity.

Keywords: *3D-printing; Rheological properties; Amplitude sweep; Viscosity*

1 Introduction

3D-printing belongs to additive manufacturing processes. Material is added layer by layer, with a predefined thickness, to create complex free-form structures (Godoi, Prakash & Bhandari, 2016). The advantage of this technology is that it is possible to produce customized products. Nowadays, it is possible to print different materials like plastics, building material, ceramics, metal and food by a 3D-printer (Fastermann, 2012). Personalised nutrition is getting more and more important (Daniel, Klein & analysiert die Varianz, 2016). By producing food with a 3D-printer it is possible to find solutions to meet the individual needs of each customer. Therefore, it is possible to customise the content of vitamins, minerals, protein and fat in the food. In order to print food, the food must be available as a homogenous mass. The rheological properties of this mass define the printability (Liu, Zhang, Bhandari & Yang, 2018). By adding connective tissue to a printable mass, the rheological properties will change. So it is necessary to

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check rheological properties such as storage modulus (G') and loss modulus (G") before printing starts. Liu, Zhang et al. (2018) printed mashed potatoes with various contents of starch. They found that added starch influences the rheological properties and also the printability (Liu, Zhang et al., 2018). Also the flow behaviour is an important factor in 3D-printing (Wang & Shaw, 2005). If one conveys product through a nozzle the shear rate will increase as the pipe diameter decreases. The viscosity will also increase if the fluid is dilatant. This makes it difficult to print dilatant products such as those with a high content of starch. The influence of rheological properties on 3D-printing has been extensively studied (Avery et al., 2014; Shao, Chaussy, Grosseau & Beneventi, 2015; Zhang et al., 2015). However, the literature and current knowledge about the influence of rheological properties on the printability of food masses are very scarce. The aim of this research was to investigate the influence of raw meat content on the rheological properties and the printability of minced meat. Minced meat represents a food with a high content of protein.

2 Materials and Methods

2.1 Preparation of the meat mass

To produce minced meat from chicken, deep-frozen chicken carcasses (Buckl Geflügel Ltd.) were thawed at 5 ^{o}C for 3 days in cold storage. While dissecting the chicken carcasses, the meat was grouped in two categories: the first category was lean meat with a low content of connective tissue (inside and outside filet, m. supracoracoideus and m. pectoralis) and the second category was meat with a high content of connective tissue. The meat with a high content of connective tissue came from the legs (m. iliotibialis, m. iliofibularis, m. fibularis, m. flexor perforans & perforatus), spine (m. latissimus dorsi, m. intercostales, m. serratus superficialis) and also skin and tendon. The lean meat was cut out of the chicken. The rest of the chicken was cooked in a boiler (WarmMaster E, Merten & Storck Ltd.) at 90 ^{o}C for 3 hours in a bag (PA/PE composite film) so as to break down the connective tissue by thermal energy. After cooking, the rest of the meat was deboned and minced by a moulinette (Moulinex Typ 6430, SEB Germany Ltd). The meat juice, formed during the thermal processing and caught in the bag, was also added to the cooked meat. After preparation, the meat was stored for 12 hours at 5 ^{o}C until it was processed. To produce the meat mass, the lean meat and the cooked meat with a high content of connective tissue were mixed in a meat processor (UMC5, Stephan Food Service Equipment GmbH) by adding crushed ice, yolk and sunflower oil (see Table 1). Crushed ice was added to cool down the mass while grinding. Preliminary investigation showed that yolk and sunflower oil improved the printability (data not presented). Only the two types of meat were added in various ratios (see Table 1). While grinding the meat, the wall of the Stephan UMC 5 was cooled by cold water to keep the temperature of the meat mass below 8 ^{o}C during processing. Hereafter, for better understanding, the concentration of cooked meat and raw meat as a percentage is related to the total weight of meat.

2.2 3D-Printing & rheological properties

The thick paste extruder (see Figure 1), with a nozzle diameter of 2 mm, was used with the 3D-Printer ZMorph 2.0S (ZMorph Ltd.). The printer was controlled by "Voxalizer 64bit" software. To evaluate the printability, five cubes were printed out for each batch. The temperature of the meat was measured with a digital thermometer (GTH 175/MO, Greisinger Electronic Ltd.) before and after the printing process. The average meat temperature was 20 ^{o}C. The cube form was defined by a G-Code, a document that defines xyz-coordinates for the extruder, as well as the piston moving speed and extrusion rate. The G-Code was created as part of this work. To build a cube, 6 layers were printed. Each layer had 7 parallel rows. The layers were 90 ^{o} rotated to the next layer. Before starting to print, the extruder was filled, air-free, with the meat mass. Then five cubes were printed. While printing the extrusion rate was 0.009 cm^3s^{-1}, the temperature of the heating bed was 20 ^{o}C and the nozzle

Table 1: Composition of each batch

sunflower oil [g]	10						
yolk [g]	10						
crushed ice [g]	100						
cooked meat with a high concentration of connective tissue [g]	0	150	200	250	300	350	500
raw lean meat [g]	500	350	300	250	200	150	0
concentration of raw meat in total meat [%]	100	70	60	50	40	30	0

moving speed was 2 cms^{-1}. The cubes and printing process were rated with grades from 1 to 5. The best grade was 1 and 5 was the worst. The printability and the optical impression were used for the evaluation, which included consideration of contour definition, stability and layout of the lines. While printing the rheological properties of the meat mass were checked. All the rheological tests were done in triplicate. Amplitude sweep and frequency sweep were performed with the Rheostress RS 300 (Thermo Fisher Scientific Inc.) using a 60 mm parallel plate with a gap of 2 mm. First the LVR was determined by an amplitude sweep. The measurements by a frequency test have to be within the limits of the LVR. The amplitude sweep was carried out by controlled shear stress (CS mode) of $\tau = 1$ Pa to $\tau = 1000$ Pa in 20 steps (logarithmic distribution) at a constant frequency of f = 1 Hz to determine the course of G' and the complex viscosity $|\eta^*|$. The complex viscosity stands for the flow resistance of the sample. It approximates to the dynamic viscosity because the measurement was taken at the LVR (Meichsner, Mezger & Schröder, 2003). To specify the frequency range, samples were measured between 0.1 to 100 Hz in 20 steps (logarithmic distribution) at a constant shear stress of $\tau = 10$ Pa.

3 Results and Discussion

3.1 Amplitude Sweep

The amplitude sweep shows that the yield point τ_y increased with an increase of raw meat content. The content of raw meat between 30 and 60% showed nearly identical yield points at the same level (see Figure 2). Outside of this range the yield point was reached earlier. The yield point correlated with the content of raw meat (α=0.05) for a quadratic relationship. A high content of raw meat results in a high content of non-cross-linked proteins. Non cross linked proteins were able to interconnect and act in this case as an emulsifier to absorb water (Sielaff, 1996). If the sample was able to interconnect, it had a reversible behavior at higher shear rate. That caused a higher yield point and also a larger LVR. If the shear rate was above τ_y the LVR was left and the deformation started to be irreversible. This area is also called the "yield zone" (Mezger, 2016). At the LVR-Plateau, the elastic modulus (G') increased with an increase of raw meat content (see Figure 3) (α=0.05). The higher the concentration of raw meat the higher the elastic modulus (G'). A high G' indicated a high degree of interconnectedness and a higher content of bound water in the meat. Only non coagulated proteins were able to interconnect and to emulsify water (Sielaff, 1996). The content of non-coagulated proteins was higher in raw meat. When proteins were able to connect they form a network. This generated network increased the ability to absorb mechanical stress, and therefore G' was higher. In meat with a high content of connective tissue, all proteins were denatured and not able to connect to each other.

3.2 Frequency Sweep

The results of the Frequency Sweep also showed that G' depends on the content of raw meat. It was difficult to distinguish the course of G' because the gradient of each curve was different

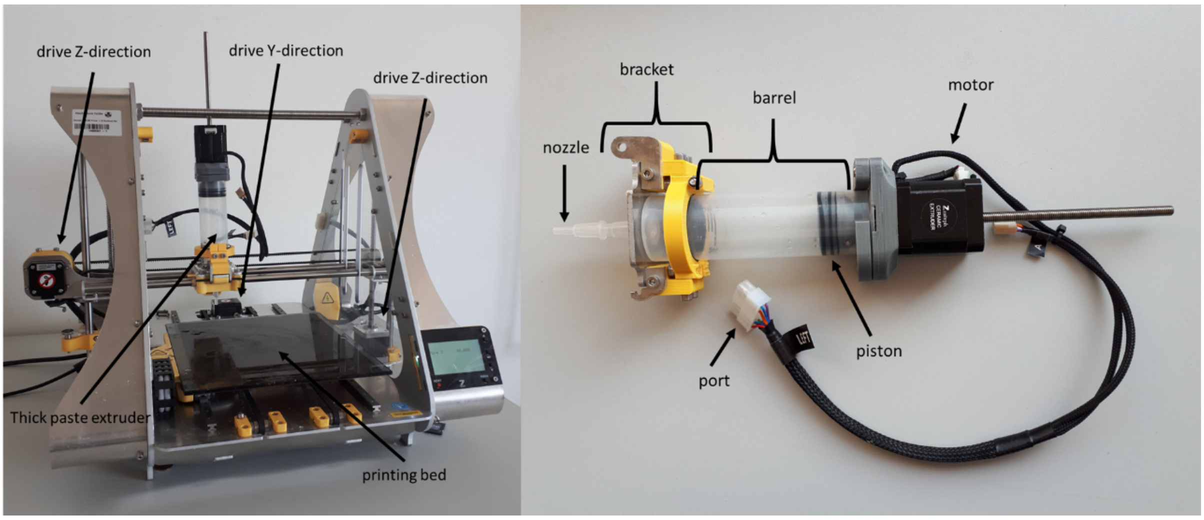

Figure 1: Zmorph 3D-Printer (left), with all the components, and thick paste extruder (right)

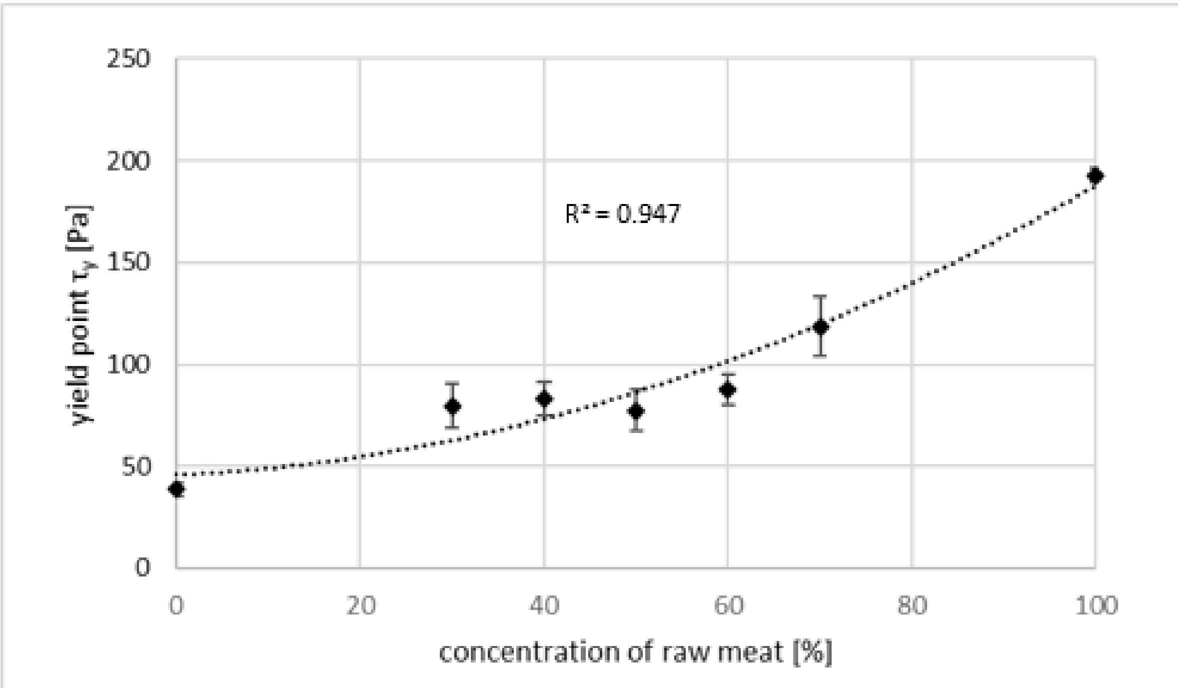

Figure 2: Yield point dependence on the concentration of raw meat, with error bars for the standard error of the mean (triple determination)

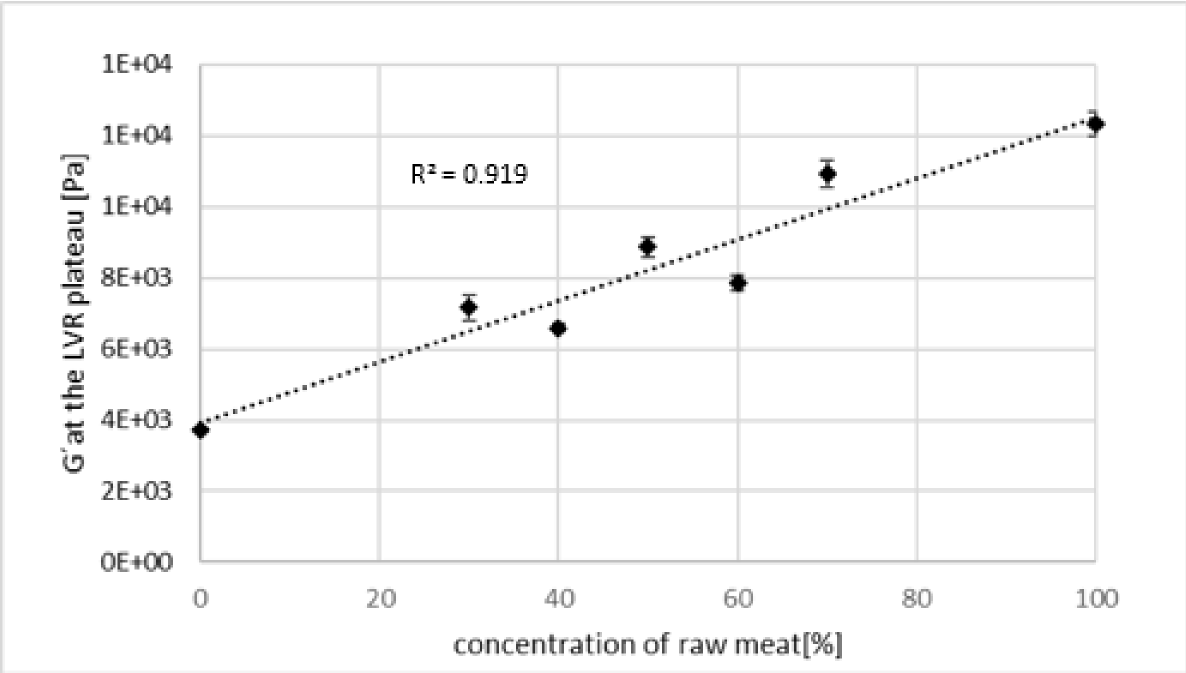

Figure 3: G' at the LVE plateau dependence on the content of raw meat, with error bars for the standard error of the mean (triple determination)

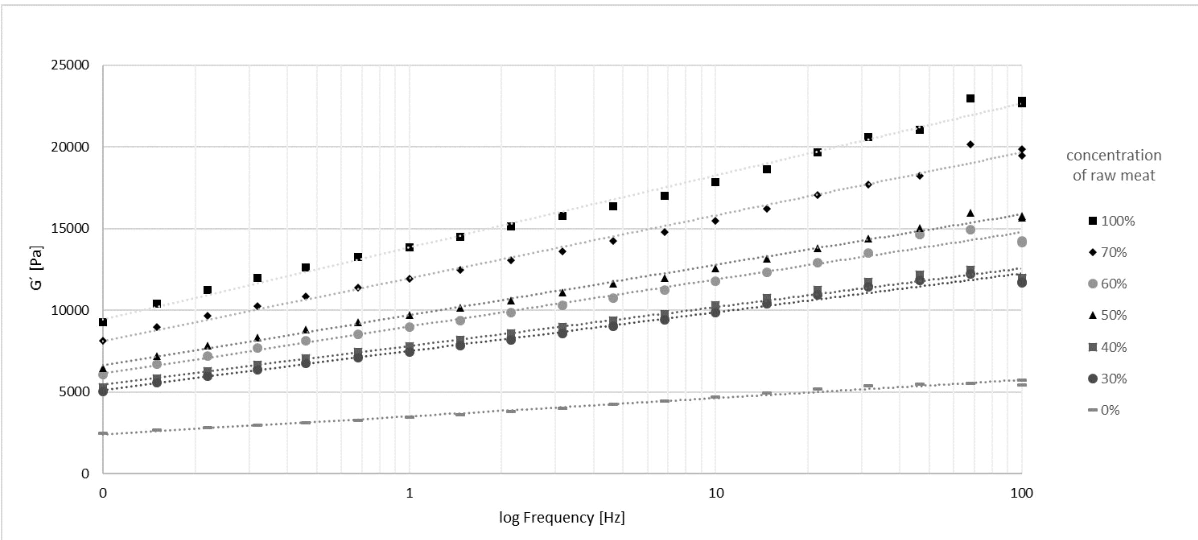

Figure 4: G' dependence on frequency at various contents of raw meat

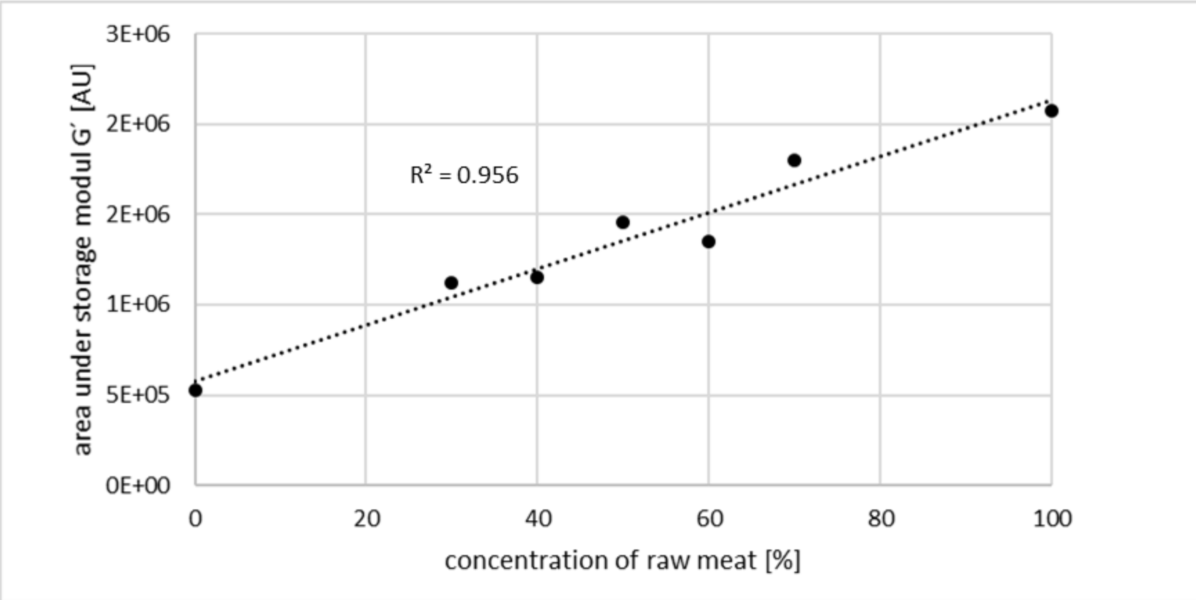

Figure 5: Area under storage modul dependence on the content of raw meat

concentration of raw meat	0 %	30 %	40 %	50 %	60 %	70 %	100 %
marks for printability	4.5	3	2	1.5	1	1.5	1
marks for optical impression	4.5	3	2	1.5	1	1.5	1

Figure 6: Pictures of printed cubes with corresponding marks for printability and optical impression at different concentrations of raw meat

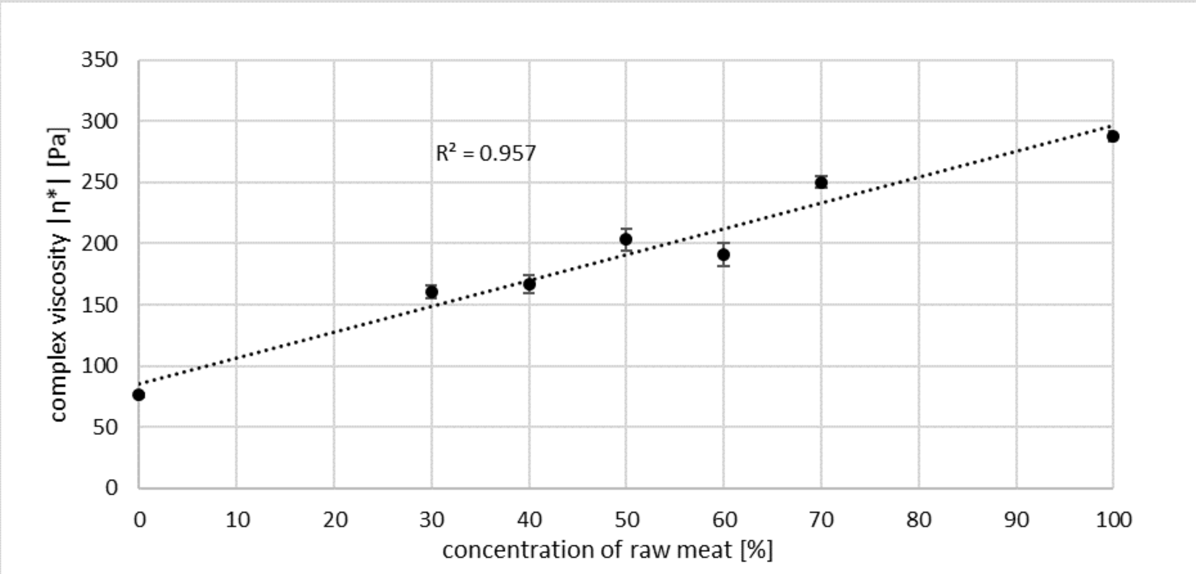

Figure 7: Complex viscosity dependence on the content of raw meat, measured at shear stress τ = 10 Pa and frequency f = 10 Hz, with error bars for the standard error of the mean

(Figure 4). The method of choice was to integrate each line and correlate the area with the concentration of raw meat. It correlates with R^2=0.956 (α=0.05) (Figure 5). The area under G' increased with a higher content of raw meat. High G' also stood for a higher degree of intermolecular connections. Proteins were able to build connections to each other through hydrogen bonds, Van-der-Waals-forces and hydrophobic effects. After heating the proteins this ability was lost as they would move freely (Ebermann & Elmadfa, 2008).

3.3 3D-printing

Printability and optical impression were improved with a higher content of raw meat. The achieved results were acceptable at >50% of raw meat. The higher the content of raw meat, the finer the printed lines and the more delicate the appearance. Also the surface was smoother at higher raw meat content. More research is needed to understand the relationship between raw meat content and the optical impression of the surface. A low content of raw meat (0% - 30%) induced an unstable form, where the cubes became wider at the base (see Figure 6). The fibres in the denatured meat gave the product an irregular surface. Generally, it can be said that with higher G' printability and optical impression are improved because of the higher content of non-cross-linked proteins in the raw meat (Rezler, Piotrowska, Dolata & Wojciechowski, 2007). The cubes with a content of raw meat above 50% were able to build a protein network. The meat would rest for 12 hours after production so that the proteins had enough time to build intermolecular connections. These connections increased the stability and also the storage modulus G'. Figure 7 shows the course of the complex viscosity $|\eta^*|$ dependence on the content of raw meat. The higher the content of raw meat the higher the viscosity ($\alpha = 0.05$). A high viscosity caused a high stability because the printing mass was prevented from flowing away during and after the printing process (Liu, Ho & Wang, 2018). The desired shape could be printed and kept.

4 Conclusion

This investigation shows that the rheological properties had a significant influence on printability and stability of printed products. The G' at the LVR-Plateau should be higher than 7000 Pa. The complex viscosity $|\eta^*|$ should be higher than 170 Pa at a shear stress $\tau = 10$ Pa and frequency f = 10 Hz to ensure sufficient firmness. The printability increased with the content of raw meat. The content of non-denatured proteins is sufficient to enable dimensionally stable printed cubes at a raw meat content above 50%.

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Production and Characterization of Emulsified Fish Mortadella From Nile Tilapia (*Oreochromus niloticus*)

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Abstract

This study produced fish mortadella from Mechanically Separated Meat (MSM) of Nile tilapia added with animal fat. Three formulations were developed: M1 (MSM - 89 % and 5 % pork fat), M2 (MSM - 84 % and 10 % pork fat) and M3 (MSM - 79 % and 15 % pork fat). The elaborated products were tested for technological, physical, physico-chemical, microbiological and sensory parameters. The results showed that the fish mortadella were microbiologically stable with a particular texture for an emulsified meat product, attractive colour and characteristic flavour. All formulations met the expected identity and quality requirements. They also achieved good acceptance by the judges, in which formulation M1 may be highlighted for presenting an emulsion stability of 97 %, higher protein content (18.09 %) and lower lipids (16.31 %). In addition, it also reached higher mean scores for texture attributes and purchase intent. Therefore, it is possible to prepare fish mortadellas from tilapia MSM using less animal fat.

Keywords: Fish waste; Fish mortadella; *Oreochromus niloticus*; Sensory analysis; Waste processing

1 Introduction

Fish are a highly beneficial food to human nutrition as they are a source of vitamins and minerals, and contain a high proportion of long chain polyunsaturated fatty acids and all the essential amino acids for humans (Fernandes et al., 2014; Sleder et al., 2015; Zotos & Vouzanidou, 2012). The Nile tilapia (*Oreochromis niloticus*) can be

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highlighted as being among the most cultivated species in the world. It has high productivity, good adaptation to diverse environments, good acceptance in the consumer market, reduced fat content and excellent fillet yield (FAO, 2017; Liu et al., 2015; Olopade, Taiwo, Lamidi & Awonaike, 2016). On the other hand, the mechanical filleting process generates approximately 70 % by-products (processing waste or discarded meat), resulting in large fish meat and oil losses. The head, carcass and guts constitute 54 % of the waste, while the skin represents 10 %. In general, these residues are destined to produce tilapia flour and oil used in manufacturing animal feed for fish, pigs and birds (Campagnoli de Oliveira Filho, Netto, Ramos, Trindade & Macedo Viegas, 2010).

Several studies have been carried out with tilapia and its by-products to promote and improve the sustainability of the fish industry, seeking to increase its consumption and maximize its use. One of its by-products is Mechanically Separated Meat (MSM), which has been used in developing new products such as emulsified meat products (sausages or mortadella, nuggets, surimi, fishburgers, etc.) It also stands out for having good unsaturated fat content which can reduce the addition of animal fat without causing sensory changes to the texture, flavour or consistency of the elaborated products (dos Santos Fogaca, Otani, De Gaspari Portella, Alves dos Santos-Filho & Sant' Ana, 2015; Ferreira Silva Bartolomeu et al., 2014).

In view of the recognized technological and market potential of emulsified meat products, the present study had the objective of producing an emulsified fish mortadela from Nile tilapia (*Oreochromus niloticus*) MSM added to different animal fat contents, aiming to obtain a differentiated product with high biological value and low fat content.

2 Materials and Methods

2.1 Tilapia slaughter and MSM production

The fish were obtained from an association of tilapia farmers from the city of Bananeiras/PB, Brazil. They were purged in clean water for 24 hours, captured in the early hours of the morning, then transported in plastic boxes where they were desensitized by thermal stunning. After respiratory activity ceased they were hung on hooks, where they were then eviscerated, and the fish were flaked, washed and filleted following a linear flow. Next, the carcasses were crushed using a pulping machine (Bresi®, 60 kg/h, 1/2, hp motor, stainless steel), thereby obtaining the MSM. The MSM was then vacuum packed into 2.0 kg polyethylene bags, labelled, and frozen (at -18 ^{o}C) until it was used in the production of fish mortadella.

2.2 Nile tilapia (*Oreochromis niloticus*) MSM quality assessment

Acidity, pH, moisture, dry matter, ash, protein (AOAC, 2012) and lipid (Floch, 1957) parameters were evaluated. The microbiological quality of the MSM was verified based on coliform counts at 45^{o}C MPN/g, coagulase-positive Staphylococcus CFU/g, sulphite-reducing *Clostridium sp.* CFU/g, *Salmonella sp.*/25 g and molds and yeasts (APHA, 2001).

2.3 Preparation of fish mortadella

The fish mortadella was prepared as described by Ferreira Silva Bartolomeu et al. (2014) and Campagnoli de Oliveira Filho, Netto et al. (2010). The present study used different concentrations of tilapia MSM and refined pork fat (Table 1). The other ingredients used in preparing the fish mortadella were added in the same concentrations to all treatments (Table 1).

A high-yielding tabletop cutter (METVISA®, CUT 2.5 L; 1/3 hp motor) was used for preparing the fish mortadella according to the flowchart and the steps described below (Figure 1).

Comminution began with the MSM, salt, curing agents and polyphosphate for thirty seconds. Ice water, pork fat, and other ingredients were added at thirty-second intervals until the formation of a paste (similar to a pâté), at a controlled temperature with a skewer thermometer (INCOTERM® Digital thermometer) with a

Table 1: Formulations of emulsified fish mortadella with different concentrations of tilapia MSM and pork fat

Ingredients	**Concentrations (%)**		
	M1	**M2**	**M3**
MSM	74.00	69.00	64.00
Pork lard	5.00	10.00	15.00
ISP	5.00	5.00	5.00
Ice/cold water	8.00	8.00	8.00
Polyphosphate	0.30	0.30	0.30
Curing salts	0.20	0.20	0.20
Antioxidant	0.60	0.60	0.60
Starch	4.00	4.00	4.00
Mortadella seasoning	0.70	0.70	0.70
Flavour enhancement	0.15	0.15	0.15
Black pepper	0.05	0.05	0.05
Garlic powder	0.25	0.25	0.25
Smoke aroma	0.50	0.50	0.50
Salt	1.25	1.25	1.25
Total	1	1	1

MSM - Mechanically Separated Meat;
ISP - Isolated Soybean Protein;
M1 - Fish mortadella with 5% pork fat;
M2 - Fish mortadella with 10% pork fat;
M3 - Fish mortadella with 15% pork fat

maximum temperature of 12 ^oC at the end of comminution.
The obtained pâté was packed into artificial shrinkable casings using a manual stuffer (METVISA®, EL.10). The fish mortadella was cooked in moist heat until it reached an internal temperature of 72±1 ^oC, monitored with the aid of a thermometer equipped with a thermocouple, Digital Cooking Thermometer). After the cooking was finished, the mortadella was submitted to thermal shock by immersing it in ice water for 15 minutes until it reached temperatures between 15 to 20 ^oC, and then it was vacuum packed.

2.4 Evaluation of technological properties

Emulsion stability - ES

Emulsion stability was performed according to the method described by Parks and Carpenter (1987). Emulsion stability values were obtained by calculating the weight loss and its percentage using the following formula:

$$\%\ Emulsion\ stability = 100 - \%loss \qquad (1)$$

Texture profile analysis

Hardness, springiness, cohesiveness, chewiness, fracturability, gumminess, adhesiveness and resilience parameters, were evaluated using the TA XT-2i texture analyser with Exponent® software (Stable Micro Systems, Godalming, UK) with a probe of 35 mm diameter (SMS P/36R) moving at a constant speed of 0.8 mm/s, according to Bourne (2002).
A Warner Braztler accessory (3 mm thick) was used to measure shear force at a test speed of 200 mm/min (Andres, Garcia, Zaritzky & Califano, 2006). Mean and standard deviation were calculated from 16 determinations (Lin & Chao, 2001).

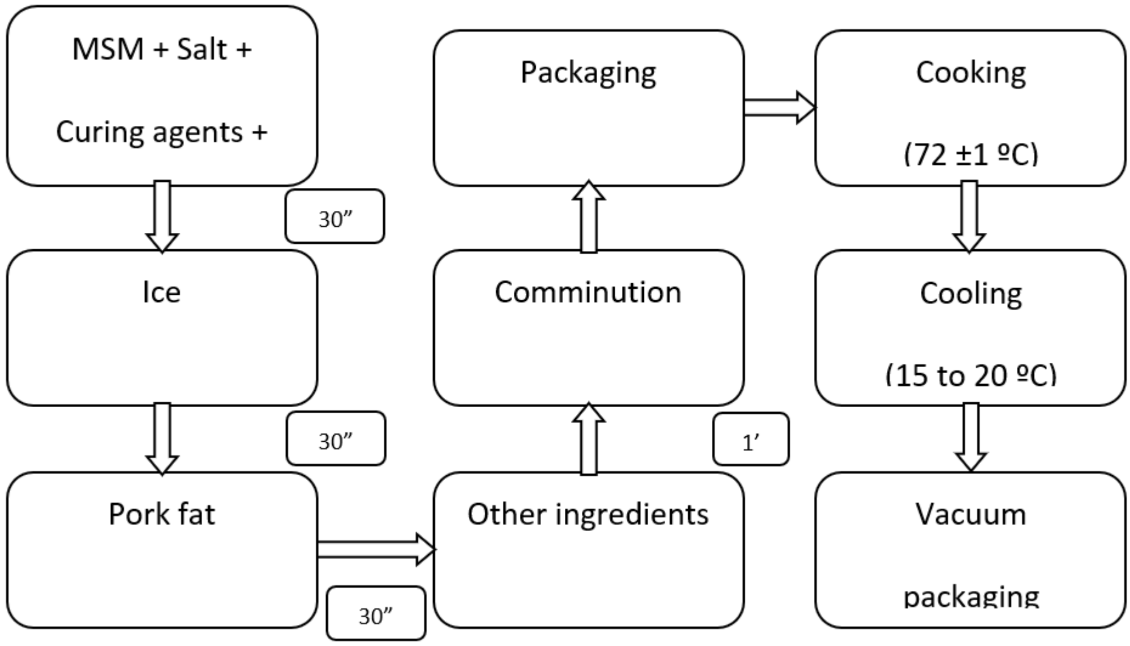

Figure 1: Flowchart of fish mortadella preparation

Physical and physical-chemical analyzes

The analyses carried out were: water activity (a_w), pH, normal acidity, moisture, dry matter, protein, ash (AOAC, 2012), chloride according to the Mohr Method, calcium content determined by EDTA volumetry (AOAC, 2012) and lipids (Floch, 1957). Water Retention Capacity (WRC) was evaluated according to the methodology by Grau (1953).

Lipid oxidation was evaluated by determining the peroxide index method (mEq/kg) based on the measurement of thiobarbituric acid reactive substances (TBARS) according to Ganhao, Estevez and Morcuende (2011) and based on Brasil (1999). The peroxide concentration was given by mEq/kg of the sample.

The instrumental colour of each fish mortadella was determined according to the methodology described by Abularach, Rocha and de Felício (1998), using a digital Minolta® colorimeter (Model CR-300, Minolta, Osaka, Japan). Luminosity parameters (L*), green (-)/red (+) (a*) and blue (-)/yellow (+) (b*) were determined according to the specifications of the *Commision Internationale de L'éclairage* (CIE, 1986).

Microbiological analyzes

Coliform counts at 45°C MPN/g, coagulase-positive *Staphylococcus* CFU/g, sulphite-reducing *Clostridium* CFU/g, and checking for *Salmonella sp.*/25 g counts were carried out as recommended for fish mortadella and for counting molds and yeasts (APHA, 2001).

Sensorial analysis

After approval of the project by the Research Ethics Committee under number 821.481, the fish mortadella was submitted for sensory acceptance, intention to purchase and order preference (Faria & Yotsuyanagi, 2002; Meilgaard, Carr & Civille, 1991; Stone & Sidel, 1993).

One hundred twenty-six (126) untrained consumers aged between 20 and 50 years of age who liked to consume fish and emulsified fish mortadella were recruited. These tasters evaluated

the samples in individual booths with white artificial lighting, away from noise and odours, and at previously established times.
The fish mortadella was offered sliced, refrigerated and codified, accompanied by salt crackers and a glass of water. Attributes of appearance, colour, aroma, flavour (specific mortadella flavour), flavour (specific fish flavour), texture and overall assessment were evaluated using a mixed nine-point structured hedonic scale (1 = I greatly disliked it; 9 = I liked it a lot). Next, the tasters were asked to indicate their intention to purchase, also using a mixed five-point hedonic scale (1 = I would never buy it; 5 = I would buy it). The samples were considered accepted when they obtained a mean $\geq$ 5.0 (equivalent to the hedonic term "I neither liked it nor disliked it"). An Acceptability Index (AI) was also calculated according to the equation:

$$AI(\%) = A \times 100/B \tag{2}$$

In which, A = average score obtained for the product and B = maximum score given to the product. An AI with good repercussion was considered $\geq$ 70 % (Teixeira, Meinert & Barbetta, 1987).
The overall preference for the different fish mortadella was also evaluated through the preference order test in decreasing order (from the most preferred to the least preferred).

2.5 Statistical analysis

Data were submitted to analysis of variance (ANOVA) and Tukey test at 5 % probability ($p<0.05$) for comparison of means using the STATISTICA 7.0 program (Statistica, 2005). Principal component analysis (PCA) was performed for the different groups of variables and based on the correlation matrix of these variables in order to provide graphical representation of the significant sensory attributes.
The results of sensory preference ordering tests were analyzed according to the Friedman test using the Newell-MacFarlane table (Faria & Yotsuyanagi, 2002).

3 Results and Discussion

3.1 Nile tilapia (*Oreochromis niloticus*) MSM quality assessment

The percentage values found for the MSM in the quality assessment are presented in the table 2. Regarding the pH, similar values in analyzing MSM from fish were reported by Cavenaghi-Altemio, Alcade and Fonseca (2013) of 7.10, and Calil Angelini et al. (2013) of 6.9. There is no legal parameter for the pH limit of fish MSM; however, for fresh fish a maximum value of 6.8 is imposed for this parameter (Brasil, 1952). The results of this study are justified by the fact that fish have a close to neutral pH. In addition, the processing for obtaining MSM disintegrates tissue, which exposes muscle fibres to degrading substances and may facilitate an increase in pH values, according to Pereda et al. (2005).
Moisture, ash and protein values were close to those found by Dallabona et al. (2013) (71.0 %, 1.22 % and 15.37 %), by Campagnoli de Oliveira Filho, Favaro-Trindade, Trindade, de Carvalho Balieiro and Macedo Viegas (2010) (74.45 %, 1.14 % and 12.76 %), and by dos Santos Fogaca et al. (2015) (73.87 % and 15.87 %) for moisture and protein, respectively.
The values for lipids were different from those found by Cavenaghi-Altemio et al. (2013) of 2.53 %, dos Santos Fogaca et al. (2015) of 7.62 %, and by Kirschnik and Macedo-Viegas (2009) of 2.91 %, due to the fact that in the present study, the MSM had the presence of the ventral musculature which contains the highest fat content, around 15 %.
The results complied with the parameters determined by the relevant legislation Brasil (2000), which recommends a minimum protein content of 12 % and a maximum fat content of 30 %, confirming that tilapia MSM is a good protein source, and thus demonstrating its viability for use in protein products of good biological value Rebouças, Rodrigues, de Castro and Vieira (2012).

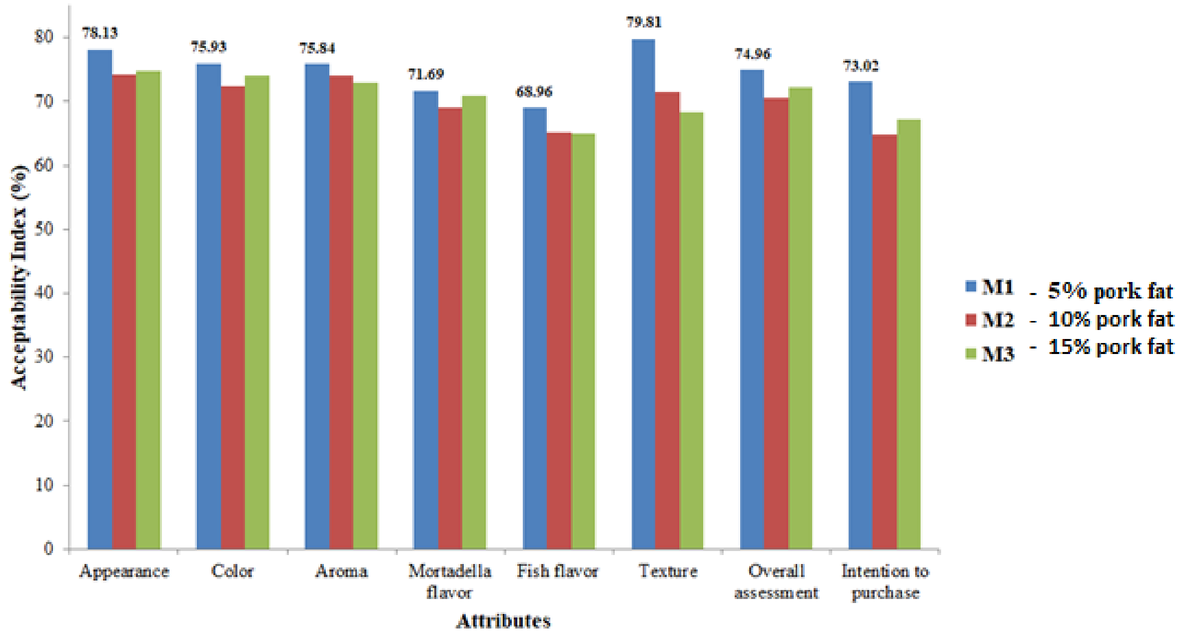

Figure 2: Acceptability index of the sensory attributes of fish mortadella with different concentrations of pork fat

Table 2: Mean values obtained in the physical and physicochemical analyses of fish mortadella with different concentrations of tilapia MSM and pork fat.

	Mortadella		
Variable	**M1**	**M2**	**M3**
a_w	0.96 ±0.00^a	0.96 ±0.00^a	0.96 ±0.00^a
pH	7.11 ±0.00^a	7.13 ±0.01^a	7.16 ±0.05^a
WRC* (%)	57.93 ±2.21^a	58.51 ±3.11^a	58.15 ±2.79^a
Normal acidity (%)	4.12 ±0.14^a	3.80 ±0.01^a	3.68 ±0.13^a
Moisture (%)	63.63 ±0.01^a	59.58 ±0.05^b	56.73 ±0.05^c
Dry matter (%)	36.37 ±0.01^c	40.42 ±0.05^b	43.27 ±0.05^a
Ashes (%)	3.90 ±0.20^a	4.31 ±0.15^a	3.85 ±0.46^a
Proteins (%)	18.09 ±0.01^a	17.46 ±0.02^b	16.54 ±0.03^c
Lipids (%)	16.31 ±0.00^c	19.84 ±0.05^b	24.38 ±0.02^a
Sodium chloride (%)	1.88 ±0.03^c	2.35 ±0.02^a	2.17 ±0.00^b
Calcium (%)	0.32 ±0.00^a	0.28 ±0.00^b	0.21 ±0.00^c

Means ± standard deviation with different letters on the same line differed by Tukey's test ($p<0.05$). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat. *WRC - Water Retention Capacity.

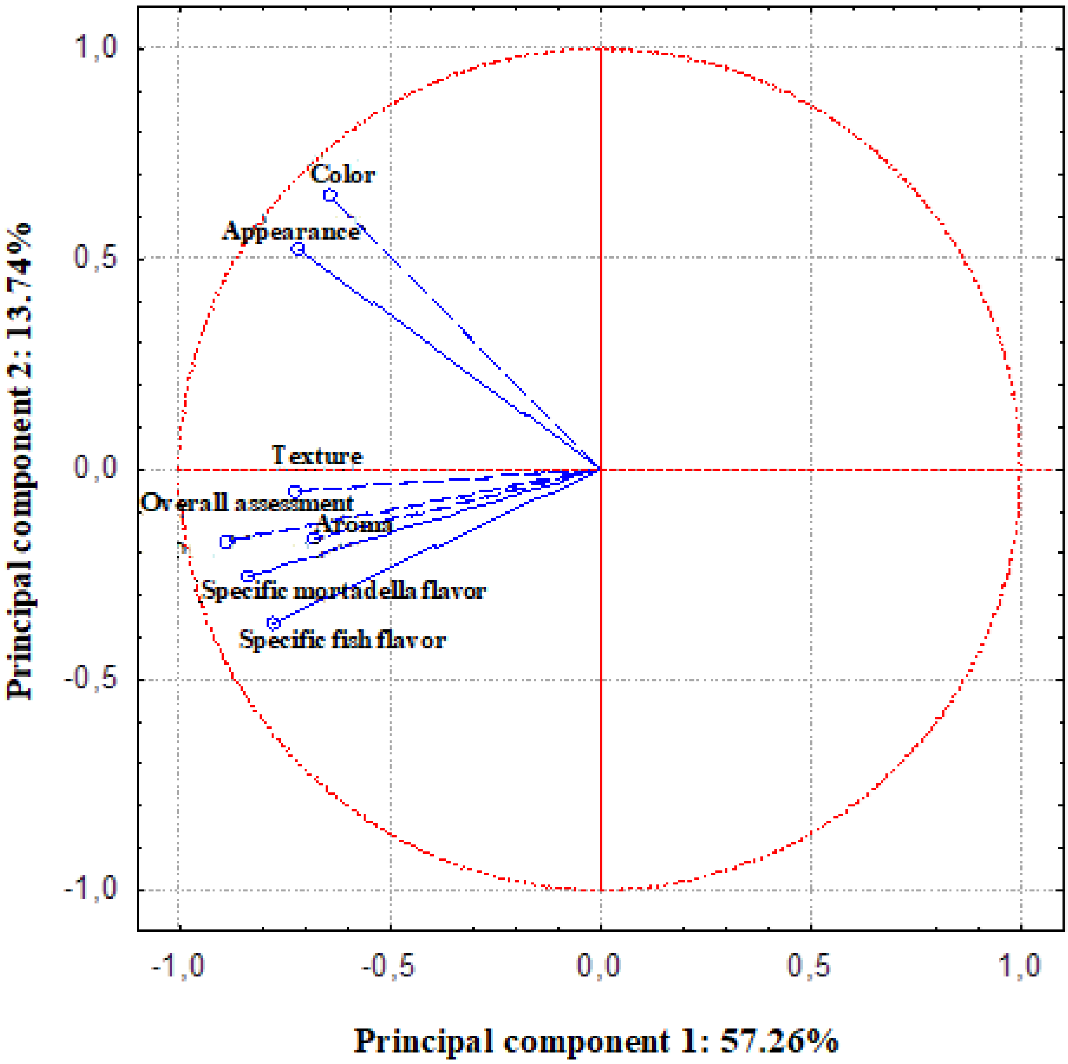

Figure 3: Projection of the PCA of the attributes in the sensory analysis of samples of fish mortadella with different concentrations of tilapia MSM and pork fat

3.2 Evaluation of the technological properties of the fish mortadella

The values obtained from the emulsion stability, shear force and texture profile evaluations are shown in Table 3.

Emulsion stability

In this study, the results for emulsion stability (ES) (97.03; 93.30 and 91.74 %) in the different fish mortadella treatments, M1, M2 and M3, respectively, were influenced by the fat content, so that the higher the pork fat concentration added, the lower the ES ($p < 0.05$). It is also important to point out that the higher amount of protein provided better emulsion stability results (Table 3) agreeing with the literature data. In evaluating sausages made from MSM and fish surimi, Yousefi and Moosavi-Nasab (2014) concluded that crude protein values of around 19% resulted in products with greater hardness and better gel consistency.
The findings of this study were attributed to the balance achieved for the protein concentrations (Table 3), which contributed to the formation of a good emulsion.

Texture profile analysis - TPA

Shear force and instrumental texture profile may vary according to the amount of water, proteins and fats found in the sample (Cengiz & Gokoglu, 2007; Cofrades, Guerra, Carballo, Fernandez-Martin & Colmenero, 2000). It was observed (Table 3) that as the MSM percentage was reduced and that of pork fat increased along with a consequent reduction in protein content, a significant decrease ($p < 0.05$) in the other parameters was also observed, with the exception of adhesiveness which increased ($p < 0.05$) with the increase in fat concentration.
Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) observed an increase in the softness of the sausages made with increasing amounts of Nile tilapia MSM - 0 to 100 %. Similarly, it was observed in the present study that the softness improved (as decreases in the hardness and shear force) with the increase in fat concentration instead of MSM. These data were expected, considering that higher amounts of fat are responsible for a reduction in texture parameters, especially when fat increases are followed by a decrease in protein and water contents in the formulations, as observed in the present study (Dincer & Cakli, 2010; Hashemi & Jafarpour, 2016; Kin et al., 2013). The results in the present study agreed with Sleder et al. (2015) who also found that a reduction in the amount of protein and an increase in the amount of fat significantly decrease the shear force from 8.4 (N) to 5.92 (N) in sausages without added fat and with 95.5 % of tambaqui meat (*Colossoma macropomum*); and with 9 % fat and 86.5 % tambaqui meat, respectively. In evaluating sausages prepared with fish MSM of fish surimi, Yousefi and Moosavi-Nasab (2014) found that those made with MSM presented greater shear force than those prepared with surimi - 1.75 N cm^2 and 1.08 N cm^2, respectively, and concluded that it is the greater amount of protein present in the MSM rather than its quality that determined the best texture profile for MSM fish sausages.
According to Hedrick, Aberle, Forrest, Judge and Merkel (1994), meat protein, represented mainly by myofibrillar proteins (actin and myosin), is the main factor responsible for the hardness in emulsified products. Colmenero, Barreto, Mota and Carballo (1995) also confirmed this relationship in their experiments with sausages, verifying that the higher the protein content, the greater the firmness of the final product; a similar characteristic was observed with the fish mortadella in this research.
Adhesiveness is high in traditional emulsified meat products (Campos, Gonçalves, Mori & Gasparetto, 1989). However, the partial or total replacement of meaty matter by MSM and the inclusion of fats contribute to forming products with a softer consistency, as in this study (Table 3). As the fat content increased in the formulations, the products had a softer consistency, tending to disintegrate during slicing.

Instrumental colour analysis

The parameters L* (luminosity), a* (red/green colour intensity) and b* (intensity of yellow/blue

Table 3: Mean values obtained in the emulsion stability, shear force and texture profile (TPA) analyses of fish mortadella with different concentrations of tilapia MSM and pork fat.

Technological parameters	Mortadella M1	Mortadella M2	Mortadella M3
Emulsion stability (%)	97.03 ±0.00^a	93.30 ±0.00^b	91.74 ±0.00^c
Shear force (N)	3.73 ±0.01^a	2.47 ±0.02^b	1.25 ±0.02^c
Texture profile (TPA)			
Hardness (kg)	1.87 ±0.06^a	1.20 ±0.04^b	0.82 ±0.01^c
Springiness (mm)	0.80 ±0.03^a	0.67 ±0.04^b	0.49 ±0.08^c
Cohesiveness (kg)	0.53 ±0.01^a	0.47 ±0.01^b	0.35 ±0.02^c
Chewiness (g.mm)	0.81 ±0.08^a	0.41 ±0.03^b	0.22 ±0.02^c
Fracturability (kg)	2.03 ±0.02^a	0.94 ±0.03^b	0.61 ±0.04^c
Gumminess (kg)	1.01 ±0.06^a	0.58 ±0.08^b	0.37 ±0.06^c
Resilience (kg)	0.35 ±0.07^a	0.21 ±0.01^b	0.11 ±0.05^c
Adhesiveness (kg.s)	-0.78 ±0.06^c	-0.34 ±0.07^b	-0.12 ±0.08^a

Means ± standard deviation with different letters on the same line differed by Tukey's test (p<0.05). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

colour) are shown in Table 4.
Colour is correlated with the acceptance of food products (Dincer & Cakli, 2010; Ferreira Silva Bartolomeu et al., 2014). It was observed that by decreasing the MSM content and increasing the fat content, the luminosity value (L*) was increased between samples M1 and M2, as well as between M1 and M3 ($p < 0.05$), confirming the theory that moisture and lipid contents have a considerable influence on L* values. The L* results were below those determined by Moreira, Visentainer, de Souza and Matsushita (2001), who found an L* value of 72.28 in a study with sausage formulated with tilapia filets, and close to those found by do Amaral et al. (2015), who also verified that the fat content positively affected luminosity indices in pork sausages as the fat content increased with values between 55 and 63.
Dantas Guerra et al. (2012) also found that the higher the fat content, the higher the L* and b* values, with values between 52.0 and 59.4 (L*) and 8.6 to 10.5 (b*) for sheepmeat mortadella prepared with different levels of pork fat ranging from 10 to 30 %, respectively. Similarly, differences were also found ($p < 0.05$) in the present study for the b* parameter when the fish mortadella with 5 % pork fat was compared with the other formulations; and the fish mortadella with 10 and 15 % fat in the formulation presented higher values for yellow colour intensity due to the higher fat percentage added to them.
There was no observed decrease for the a* values when the amount of MSM also decreased. This occurred due to the fact that we chose to use isolated soybean protein to improve the acceptance of the fish mortadella, which probably contributed to an increase in the red colour between the M1 fish mortadella and the other formulations ($p < 0.05$).

3.3 Evaluation of physical and physical-chemical characteristics of fish mortadella

The physical and physico-chemical parameters of the fish mortadella are shown in Table 2.
The results of water activity (a_w) obtained for the fish mortadella were equal ($p > 0.05$), corresponding to 0.96 and above 0.85, which is the

Table 4: Mean values obtained in the instrumental color analyse of fish mortadella with different concentrations of tilapia MSM and pork fat.

	Mortadella		
Technological parameter	**M1**	**M2**	**M3**
Instrumental Colour			
L*	62.44 ±0.26^b	65.21 ±0.13^a	65.31 ±0.09^a
a*	9.57 ±0.25^b	10.23 ±0.17^a	10.76 ±0.31^a
b*	10.76 ±0.06^b	11.21 ±0.16^a	11.35 ±0.05^a

Means ± standard deviation with different letters on the same line differed by Tukey's test ($p<0.05$). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

limit for considering them as foods with high water activity. This characteristic favours the proliferation of microorganisms, indicating the need to keep these products under refrigeration during storage, as well as the adoption of other measures during processing to inhibit microbial proliferation. These results are similar to those established by Fellows (2009) for a cooked cured product (0.95); by Ferreira Silva Bartolomeu et al. (2014) who determined a_w of 0.98 for a smoked tilapia mortadella; and to the values reported by Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) of 0.98 for tilapia MSM sausage; and also those by Dallabona et al. (2013) corresponding to 0.97-0.98 for tilapia MSM smoked sausages under different heat treatment conditions and packaging.

The pH values were similar to the results obtained for MSM. Other researchers such as Sleder et al. (2015) reported values lower than those found in this study, with mean pH values of 6.6 in tambaqui sausages (*Colossoma macropomum*); Cavenaghi-Altemio et al. (2013) determined mean values of 6.76 in analysing sausages produced from protein concentrates of tilapia; and (Mélo et al., 2011) found mean values of 6.39 in tilapia MSM mortadella.

In the present study, the pH values can be justified since the material used was raw, which already has higher values of pH, as well as sodium polyphosphate in the formulation (Mélo et al., 2011) which has a pH between 9.5 and 10.5.

There were no significant differences in the WRC values between the different fish mortadella formulatios (Table 2). However, these results were lower than those reported by Sleder et al. (2015). These authors identified WRC values ranging between 71.83 and 74.71 % in tambaqui mortadella with different fat percentages. Despite the low WRC values in this study, there was no damage to the structure of the formed emulsion, such as fat coalescence or syneresis, and it did not affect the sensory evaluations.

Regarding to moisture content, similar results have been reported for different types of emulsified meat products with MSM. For example, Sleder et al. (2015) observed a decrease in moisture values (72.73 %, 71.05 % and 68.82 %) as the fat content in the tambaqui sausage formulation increased (0 %, 4.5 % and 9 %, respectively). The results found in this study were similar to those of Yousefi and Moosavi-Nasab (2014), who studied MSM sausages or tilapia suprimi and found moisture values of 65.46 and 67.45 %, respectively; and those of Mélo et al. (2011), who analysed the moisture of tilapia MSM mortadella and determined decreasing values between 75.90 and 59.38 % as the fat percentage varied between 0 to 20 %, a similar behaviour to that found in the present investigation.

Despite the variations observed according to the added pork fat content, all formulations prepared in this study remained within the limits of moisture recommended by legislation Brasil (2000), which is 65 %. This differed from other studies with other types of emulsified meat products in which the average moisture of their products was above those quantified here, such as Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010), who produced tilapia MSM sausages with a mean moisture content of 70.75 %; and Cavenaghi-Altemio et al. (2013), who processed emulsified meat products with concentrated tilapia protein and found average moisture values of 75.83 %.

Increasing pork fat content caused the dry matter content to increase significantly ($p < 0.05$) in all formulations (Table 2). In addition to the increasing pork fat concentration in the formulations, the values were in line with the composition of the MSM extracted from the abdominal muscle, which contained considerable adiposity (Bordignon et al., 2010; de Oliveira, Henriques Lourenco, Sousa, Peixoto Joele & do Amaral Ribeiro, 2015). It is worth mentioning that fish fat is a source of omega-3, a fatty acid beneficial to consumer health (Menegazzo, Petenucci & Fonseca, 2016; Moreira et al., 2001; Nestel, 2000).

The values for ash represented the overall mineral content of the formulations, which did not differ significantly between them ($p > 0.05$) despite the fat percentage and MSM variations. Similar results were found by Sleder et al. (2015), who found quantities between 3.86 and 4.20 % in tambaqui sausages with different fat contents; by Dallabona et al. (2013), who found values between 3.44% in pasteurized sausage and 3.97 % in smoked sausage; and by Campagnoli de Oliveira Filho, Netto et al. (2010), who obtained similar results (3.40 %) in cooked emulsified meat products. The high levels of ash occurred due to the addition of ingredients such as salt and additives, as well as the incorporation of bone fragments to the mass.

The protein and fat content of the fish mortadella varied significantly ($p < 0.05$). Consequently, a concomitant decrease in protein content and increase in lipid content was observed for M1, M2 and M3. The importance of these nutrients and their equilibrium in forming a good emulsion and maintenance of their shelf life stability is known, in addition to the importance of the amount of proteins for greater firmness of the formed gel (Yousefi & Moosavi-Nasab, 2014). Thus, having a balanced percentage in the amount of proteins and fats is of fundamental importance to ensure obtaining a product with all the desirable characteristics present.

The fish mortadella formulated in the present study are products of high biological value and with reduced fat content, in addition to being a source of unsaturated fat. This is especially true for the M1 formulation, which presented a percentage of 18.09 % for proteins and 16.31 % for fats (almost half of that recommended by the legislation, which is at most 30 %). These results reinforced the relevance of this new product being proposed for the food industry.

In relation to sodium chloride, a statistical difference ($p < 0.05$) between the treatments was observed (Table 2), probably due to the fact that the processing is semi-industrial and carried out in batches of 500 g each, which may have caused differences in the homogenization pattern of the ingredients.

Calcium levels are well below that recommended by the legislation of a maximum of 0.90 % (Brasil, 2000). As expected, it was also observed that calcium values decreased ($p < 0.05$) as MSM was replaced by animal fat in the formulations. This fact is due to the reduction of the tilapia filleting residues, which contain bone fragments in their composition (calcium source).

Although there are no legal standards limiting the levels of lipid oxidation expressed as peroxide value, values above 1.51 mg of mEq/kg and 3.0 mEq/kg in fish and fish products, respectively, are classified as unacceptable (Campagnoli de Oliveira Filho, Favaro-Trindade et al., 2010). Thus, it is essential to verify the presence of oxidation in emulsified meat products through an evaluation of the peroxide index (mEq/kg), considering that it is possible to have important alterations in the food depending on its presence in the product due to lipid oxidation.

No changes to this parameter were found in this study with results of 0.0 mEq/kg, which showed that all the processing was in accordance with what is recommended by Good Manufacturing Practice (Brasil, 1997), and unlike other studies such as Ferreira Silva Bartolomeu et al. (2014), who found levels of 0.99 mEq/kg in tilapia mortadella; Calil Angelini et al. (2013), who quantified a mean of 0.22 mEq/kg in tilapia quenelles; and Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010), who found levels between 0.67 mEq/kg and 1.24 mEq/kg in sausages with percentages of tilapia MSM ranging from 0 to 100 %.

3.4 Microbiological evaluation of MSM and fish mortadella

All the formulations were in accordance with RDC No. 12 of January 2001 - National Health Surveillance Agency (Brasil. Agência Nacional de Vigilância Sanitária. Collegiate Board Resolution No. 12 of 2001. Approves the technical regulation on microbiological standards for food. Diário Oficial da República Federativa do Brasil, Poder Executivo, Brasília, DF, Brazil (in Portuguese)., 2001), and no viable cells were found for any of the microorganisms that were tested for. These results agree with those found by Ferreira Silva Bartolomeu et al. (2014), who formulated smoked tilapia MSM mortadella; and those of Campagnoli de Oliveira Filho, Netto et al. (2010) and Dallabona et al. (2013), who also elaborated tilapia MSM mortadella and tested its microbiological quality. These findings affirmed the feasibility of producing this type of emulsified meat product.

3.5 Sensory evaluation

Only the attributes texture and intention to purchase differed significantly among the treatments ($p < 0.05$) (Table 5). This occurred due to the correlation of the data for the instrumental texture and sensory texture. The increase in the fat content in the formulations corresponded to a reduction in the instrumental texture, which probably contributed to the reduction in the scores attributed to sensory texture ($p < 0.05$).

Nonetheless, all formulations presented mean scores equal to or above 6.0 (Table 5), demonstrating that the fish mortadella was sensorially well-accepted for this parameter; a behaviour also similar to that found by Rahman, Al-Waili, Guizani and Kasapis (2007), who observed a correlation between instrumental and sensory texture in fish sausages.

A positive correlation between instrumental hardness and fracturability with the sensory texture was observed, although they were higher for the M1 formulation (Table 3). There was good sensory acceptance for the texture attribute, presenting a higher mean score of 7.3 ($p < 0.05$) when compared to the other formulations. These findings were similar to the data found by Rahman et al. (2007), which showed that instrumental hardness is highly correlated with the sensory hardness of fish sausage.

The sensory scores for colour, odour, flavour and texture were higher than from studies with other emulsified products such as by Campagnoli de Oliveira Filho, Netto et al. (2010) and Cavenaghi-Altemio et al. (2013). In the present study, scores were similar to the results of Ferreira Silva Bartolomeu et al. (2014), who evaluated smoked tilapia MSM mortadella and found scores ranging from 7 (I moderately liked it) to 8 (I liked it a lot).

In addition, despite being a new product to the consumers, it was found that their overall acceptance varied between the hedonic terms “I slightly liked it” and “I liked it a lot”, with a mean score ranging from 6.8 ($\pm$1.5) to 7.1 ($\pm$1.4). Campagnoli de Oliveira Filho, Favaro-Trindade et al. (2010) found lower results than these for the overall assessment of sausages prepared with increasing percentages of MSM (0 to 100 %), with scores ranging from 6.1 to 5.0.

In an emulsion, the lack of balance between the amounts of fat and protein can harm its sensory evaluation (Yousefi & Moosavi-Nasab, 2014), which was not observed in this study considering that all formulations generally obtained average scores for the sensory attributes between 6 and 7. The M1 formulation stood out with scores for the appearance attributes, mortadella flavour, texture and overall assessment between 7 and 8, whose hedonic terms correspond to “I moderately liked it” and “I liked it a lot”.

These scores were reinforced by the comments made by the judges of which 105 out of the 126 tasters made some kind of comment with an emphasis on taste and texture attributes, with about 32.38 % and 10.47 % reporting that the samples presented “pleasant taste” and “pleasant texture”, respectively. The other comments were basically summarized as: “strong and/or smoky flavour” (13.33 %), “fishy taste” (7.61 %), “pleasant smell” (5.7 %), among others.

Other important data relates to the purchase intention, whose attributed scores ranged from the hedonic terms of 3 (“I would perhaps buy

Table 5: Mean scores of the sensory acceptance and purchase intention tests performed with of fish mortadella with different concentrations of tilapia MSM and pork fat.

Attributes	Mortadella		
	M1	**M2**	**M3**
Appearance	7.0 $\pm$1.5^a	6.7 $\pm$1.6^a	6.7 $\pm$1.6^a
Colour	6.8 $\pm$1.6^a	6.5 $\pm$1.7^a	6.7 $\pm$1.8^a
Aroma	6.8 $\pm$1.8^a	6.7 $\pm$1.7^a	6.6 $\pm$1.7^a
Specific mortadella flavour	7.0 $\pm$1.5^a	6.8 $\pm$1.5^a	6.9 $\pm$1.5^a
Specific fish flavour	6.8 $\pm$1.4^a	6.7 $\pm$1.4^a	6.5 $\pm$1.5^a
Texture	7.3 $\pm$1.4^a	6.6 $\pm$1.7^b	6.0 $\pm$1.1^c
Overall assessment	7.1 $\pm$1.4^a	6.8 $\pm$1.5^a	6.9 $\pm$1.5^a
Intention to purchase	3.9 $\pm$1.1^a	3.2 $\pm$1.3^b	3.0 $\pm$1.0^c

Means $\pm$ standard deviation with different letters on the same line differed by Tukey's test ($p<0.05$). M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

Table 6: Contribution to the formation of the principal component of the attributes evaluated in the sensorial analysis of mortadella samples.

Analyzed Attributes	Principal Components	
	1	**2**
Appearance	-0.716	0.526
Colour	-0.646	0.653
Aroma	-0.681	-0.167
Specific mortadella flavour	-0.836	-0.256
Specific fish flavour	-0.776	-0.367
Texture	-0.723	-0.052
Overall assessment	-0.889	-0.168

Table 7: Distribution of grades according to the ordination of general preference by the judges (n = 126) in the sensorial analysis of fish mortadella with different concentrations of tilapia MSM and pork fat.

Mortadella	Number of Judges per Order*			Sum of orders**
	1	2	3	
M1	34	33	59	277^a
M2	55	44	27	224^b
M3	57	44	25	220^b

* 1 = least preferred, 3 = most preferred.
** Sum of the orders of each sample = (1 x number of judges) + (2 x number of judges + (3 x n^o judges). a, b - lower case letters indicate the significant differences between of the mortadellas ($p<0.05$) by the Friedman test.
M1 - Fish mortadella with 5% pork fat; M2 - Fish mortadella with 10% pork fat; M3 - Fish mortadella with 15% pork fat.

it/maybe not buy it") and 5 ("I would buy it"), with emphasis on the sample prepared with 5 % pork fat in its formulation which had a higher mean score ($p < 0.05$) when compared to the other formulations.

Figure 2 shows the acceptability indexes (AI) for the fish mortadella with different concentrations of tilapia MSM and pork fat. According to Teixeira et al. (1987), it is necessary that a product reaches an AI of at least 70 % in order for it to be considered as accepted in terms of sensory properties. Thus, it was observed that the AI for most of the attributes evaluated in the 3 fish mortadella formulations was above the minimum, with emphasis on the M1 fish mortadella.

The findings of this study agreed with Sleder et al. (2015) who verified that the best acceptance rates achieved in a study with tambaqui meat sausages were for those with lower pork fat content (9 %), with mean acceptance rates of 80 % for all the tested parameters. Mélo et al. (2011) also found expressive acceptance levels (78.43 %) for the overall assessment attribute in fish mortadella prepared with corn oil and wheat fibre.

The intent to purchase test indicated a tendency of purchasing fish mortadella with 5 % pork fat by a large proportion of the judges who participated in the sensory evaluations (73.02 %), as shown in Figure 2. Results similar to this were observed by Mélo et al. (2011), who reported an intent to purchase of 78.43 % for mortadella prepared with MSM, corn oil and wheat fibre. The data obtained in this study indicate that there is room in the market for the product when it is available, increasing the supply of nutritious and healthier products.

The principal component analysis of the attributes in the sensory acceptance analysis of the fish mortadella was important to verify which attributes most contributed to differentiating the three formulations, which is shown in Figure 3.

In the PCA, the first main component contributed to 57.26 % of the total variance and the second with 13.74 %, representing the first two factorial axes (71 %) in the total variance. If the first two or three components in a PCA accumulate a relatively high percentage of the total variance (generally above 70 %), they will satisfactorily explain the variability manifested among the samples (Mardia, 1979), and corroborate the data found in the present study.

For the Principal Component 2, colour and appearance were the attributes that most contributed to the group separation, as can be seen in Table 6. Thus, for the target audience in question who analyzed the fish mortadella formulations under study, the most important quality to differentiate them from one another were these sensory attributes, with particularly little influence by the fish taste and mortadella flavour attributes.

The preference order test allowed us to determine which product was most preferred by a particular target audience through indications of the judges, ordering samples from the "most preferred" to "least preferred". The results for the preference order are shown in Table 7.

According to the statistical analysis, a significant difference ($p < 0.05$) was observed between the M1 samples and the other formulations, in which the sample with 5 % pork fat in its constitution was more preferred, most probably due to the texture attribute since it also presented a higher score ($p < 0.05$) on the sensory acceptance test, also justifying its higher purchase intent.

When judges were asked which sensory characteristics they enjoyed the most in their preferred sample (M1), the majority of the answers were the flavour, the texture and the aroma. These attributes were similarly mentioned as characteristics that were not appreciated in the less preferred samples (M2 and M3).

4 Conclusion

In general, it can be affirmed that the different fish mortadella formulations had a good acceptability, with emphasis on the fish mortadella with 5 % of pork fat which, in addition to exceeding the minimum limits predicted to be considered approved by the consumer public, also presented excellent results for technological, physical parameters and nutritional value. The development of this study proved that technological processing of an emulsified meat product from fish capture to its consumption is feasible from the logistic, sanitary, and technological points of view, among others. The use of MSM did not depreciate the quality of the final product. We suggest that the production of this mortadella can represent a viable technological alternative for the cluse of tilapia processing waste.

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Industrial Practice for Reducing Defective Sterile Milk Products Produced Using Overpressure Rotary Retorts

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Abstract

Indonesian consumers are fond of commercially sterilized milk as indicated by increasing product sales. High demand for products intensifies the need to increase productivity, generally achieved by minimizing product defects. This study aimed to reduce the number of defects in commercially sterilized milk produced using overpressure rotary retorts. Based on Pareto analysis, the percentage of defective products was 5.14% of which 2.37% were dented bottles. A cause-effect diagram (Ishikawa Diagram) was used to find the root cause of dented bottles. The pressure difference between the retort chamber (external pressure) and inside the product packaging (internal pressure), and the number of bottles stacked inside the retort basket (bottle density) were found as major factors for causing dented bottles. The internal pressure was 1.20 bar higher than the external pressure. By reducing the pressure difference to 0.40 bar, the percentage of dented bottles could be reduced to 0.79%. Applying the lowest bottle density (73% of the retort basket area occupied by bottles) during the sterilization process could decrease the number of dented bottles, however, it also increased the appearance of striped lids. The best conditions for sterilization (pressure difference = 0.40 bar; number of bottles/basket = 1938 bottles) which were used in the three-month full-scale production trial reduced the percentage of defective products from 5.14% to 2.24% of which 0.76% were dented bottles. Setting the retort pressure at 2.80 bar could avoid 52,920 defective bottles of commercially sterilized products per month.

Keywords: Commercially sterilized milk; Product defects; Dented bottles; Overpressure rotary retorts; Sterilization

1 Introduction

Based on the Neilsen RMS 1^{st} quarter of 2019 Survey in Indonesia, consumption of liquid and powder milk increased by 11% and 5%, respectively. Increased consumption of liquid milk in Indonesia makes competition among producers even tighter. To remain competitive notably requires continuous innovation and cost reduction strategies during the production of dairy-based products (Kong, Yang & Xu, 2019; Magnusson & Berggren, 2017). In terms of improving a product's quality and productivity, reducing product defects may be considered as a strategy (Zhang, Kano, Tani, Mori & Harada, 2018). According to Ahmad and Ginantaka (2018), product defect categories in commercially sterilized bottled milk are leak, blown, nonstandard label, no coding, dented bottle, narrow seal, folded, wrinkle and overheat. The bottle used for commercially sterilized milk is mostly made of HDPE (high-density polyethylene) (Potts, Amin

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& Duncan, 2017). As an elastic material, HDPE bottles will expand when exposed to heat (Zheng, Zhang, Ma, Wang & Yu, 2019). Balancing the internal and external pressure of the packaging during the sterilization process is important to prevent deformation defects in semi-flexible packaging (Hariyadi, 2017). Primanintyo, Syafei and Luviyanti (2016) reported a successful application of the six sigma method to reduce defects in the production of car tyres. A similar approach to decrease product defects in the textile industry (Heijacker company) was also reported by Wulandari and Bernik (2018). These studies indicated that product defects could be reduced by up to 50%. This study aims to reduce the number of defective commercially sterilized milk products in HDPE bottle produced using overpressure rotary retorts.

2 Materials and Methods

2.1 Materials

High-density polyethylene (HDPE) bottles (volume of 140 mL) produced by Kalbe Milko Indonesia Company were used in this research. Commercially sterilized milk was produced according to the standard operating procedures (SOP) of the company. The sterilization process was carried out using a Stock DFT Rickerman SRX 1300-4-HV-PS overpressure rotary retort (Germany) equipped with PicoVACQ Orion TMI 1 C data logger (France) and an Ellab Track Sense Pro - Wireless Data Logger (Denmark). Statistical data analysis was carried out by Minitab 19 (United States).

2.2 Methods

Pareto Analysis

Data used for Pareto analysis was from the production period of March 2018-February 2019 at the Kalbe Milko Indonesia Company. The defect categories and number of defects were collected during this period. Based on the Pareto analysis, the defect category with the highest percentage of all defects was further investigated and discussed within this study.

Cause-effect Analysis for the Selected Product Defect Category

An Ishikawa (fishbone) diagram was used to investigate the selected product defect category. Factors causing the defect in the selected product defect category were analyzed through discussions with machine suppliers and production, engineering and utilities unit staff in the company. The cause-effect analysis covered four aspects: man, method, machine and material. The possible root causes of each aspect were determined by focus group discussions.

Improvement of the Largest Defect Category

Possible root causes obtained from the cause-effect analysis were mitigated and implemented during production. During the production trial, the heat adequacy of the sterilization process was measured in terms of minimum F_0 value. The F_0 value was determined using the trapezoidal method, based on dynamic product temperatures recorded by the data logger (Ellab Track Sense Pro Wireless Data Logger).

Verification of Improvement in Results

A 3-month production trial was carried out to verify the improvement. The improvement was quantified by comparing data from the 3-month production trial and the production for one year prior to this study covering the number of product defects in all identified defect categories.

3 Results and Discussion

3.1 Pareto Analysis of Product Defects

Pareto analysis is used to analyze a series of processes that aim to overcome weaknesses or problems that arise (Wang & Choi, 2019). Pareto analysis focuses on risk that might occur when designing and modifying a process. The Pareto analysis principle lies in determination of the 20% of main causes that must be dealt with so as to solve 80% of the problems. Absolute values can be arranged in Pareto diagrams to show which technical characteristics are the most important and the risk priorities of existing requirements (Erdil, 2019). Pareto analysis is also used to prioritize determinants that can be a focus for improvement (Shivajee, Singh & Rastogi, 2019). Pareto diagrams of defective products from March 2018 to February 2019 are shown in Figure 1. The largest contributor to total product defects was dented bottles (2.37 %). Total product defects were 5.14% of the total production. Therefore, dented bottles contributed 46.07% of the total product defects. In this study, the dented bottle defect category became the priority for improvement to identify the root cause through fishbone diagram analysis.

3.2 Cause-effect Analysis for the Largest Product Defect Category

Dented bottles were observed to occur during the sterilization production process. The determination of cause-effect in the Ishikawa's diagram is unlimited but it can be used to determine significant factors (Simanova & Gejdos, 2015). Fishbone diagrams require a focus group discussion as the main tool to identify the main problem (Elleuch, Dafaoui, El Mhamedi & Chabchoub, 2016). Through discussions with machine suppliers, production teams, engineering teams and the utility team, the main causes of the problem for each factor were found. The cause-effect analysis results are shown in Figure 2.

Man

The man factor in the cause-effect analysis is the operator who operates the retort. Retort operators' technical knowledge, technical skills and technical behavior were evaluated by experts from the machine supplier. Each production shift had one operator of the retort and there were three shifts in one day. Every year, the supplier provides refresher training for the operators and assesses their competencies. An evaluation of operators' competencies was based on pre-test and post-test results, with a minimum pass score of 8.0. Based on Table 1, the technical knowledge, technical skills and technical behavior of the operators meet the supplier's standards. Therefore, the retort operator is not the cause of dented bottles.

Table 1: Pretest and posttest scores for retort operators trained by the machine supplier.

Operator	*Pretest*	*Posttest*	Note
Operator A	8.0	10.0	Pass
Operator B	8.5	9.5	Pass
Operator C	8.0	9.5	Pass

Machine

Closed containers of the food to be sterilized are placed in the retort for a certain time, at high temperature and pressure. A commonly used material of containers in a retort process is HDPE (High-Density Polyethylene) plastic. The sterilization process used with semi-flexible packaging requires over pressure to maintain the integrity of the packaging. Packaging damage can occur due to an improper combination of increased pressure and expansion of the product in the containers (Augusto, Tribst & Cristianini, 2014). The sterilization process parameters that must be controled are temperature, time, pressure and temperature distribution in the retort (Hariyadi, 2017). The retort supplier ensures the good working condition of the retort through a retort maintenance agreement. The supplier's annual retort inspection report in June 2018 showed that the retorts were in good condition and working properly. A reading from the retort thermometer was within the tolerance limit based on the calibration results. The retort thermometer calibration was performed at temperatures between 100.00 ^{o}C and 122.00 ^{o}C, with a maximum correction of + 0.1597 ^{o}C which is below the maximum correction required by the maker (± 0.5000 ^{o}C). Other checks were carried out on the timer used in the retort, the pressure gauge of the retort, the rotary speed of the basket and the sterilization value obtained by the products. Checks on the timer were carried out from 5 minutes to 15 minutes, with the results of

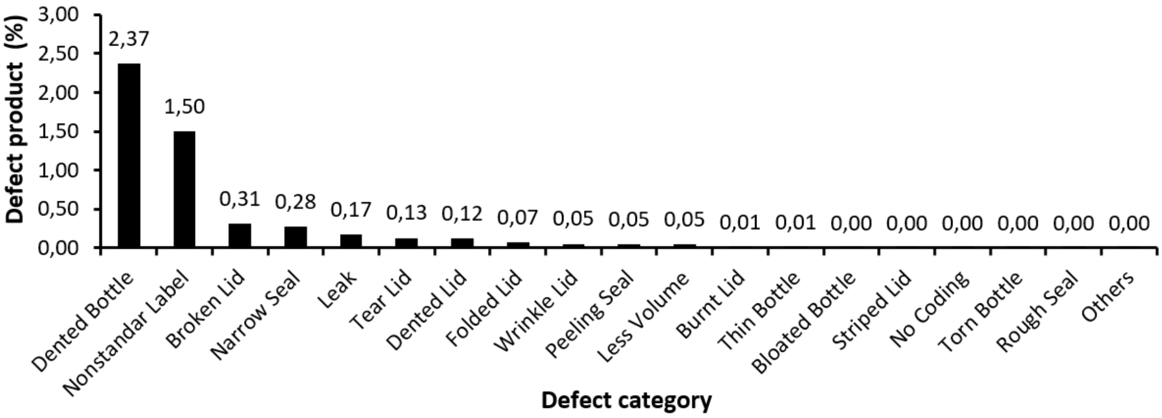

Figure 1: Pareto diagram of product defect categories during one year of production

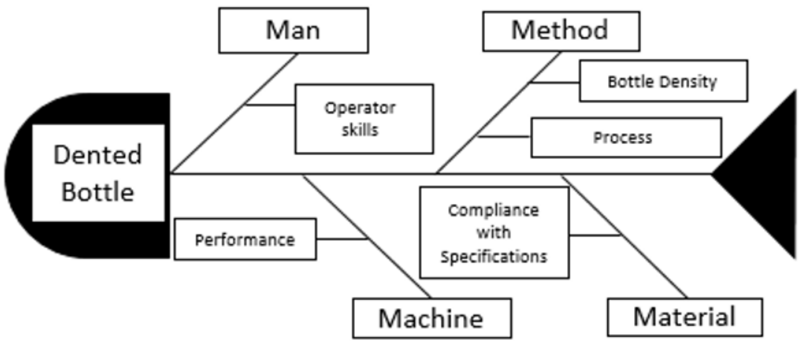

Figure 2: Fishbone diagram for the dented bottle defect category

correction at 10 minutes showing 1 second. This time correction is within the maximum correction limit required of 5 seconds. The pressure gauge calibration was carried out in the range of 1 bar to 4 bar. The correction value of the pressure gauge was + 0.011 bar which is below the maximum correction of ± 0.050 bar. The rotational speed of the basket was inspected at 800 to 1000 rpm. Correction for the rotational speed was + 5 rpm which is lower than the maximum tolerable value of ± 10 rpm. The validation of the sterilization value results ranged from 10.41 - 13.29 minutes which meet the required minimum sterilization value of 10.00 minutes. Based on these annual inspection results it can be concluded that the over pressure rotary retort is in good condition and effectively performs the required sterilization process.

Material

Material factors observed in the fishbone diagram are specifications of bottles and lids. The bottle used is a single layer type HDPE, with a colorant agent. The HDPE material functions as a radiation barrier and oxygen barrier. Inspection of bottles was carried out by comparing the required and actual specifications. Based

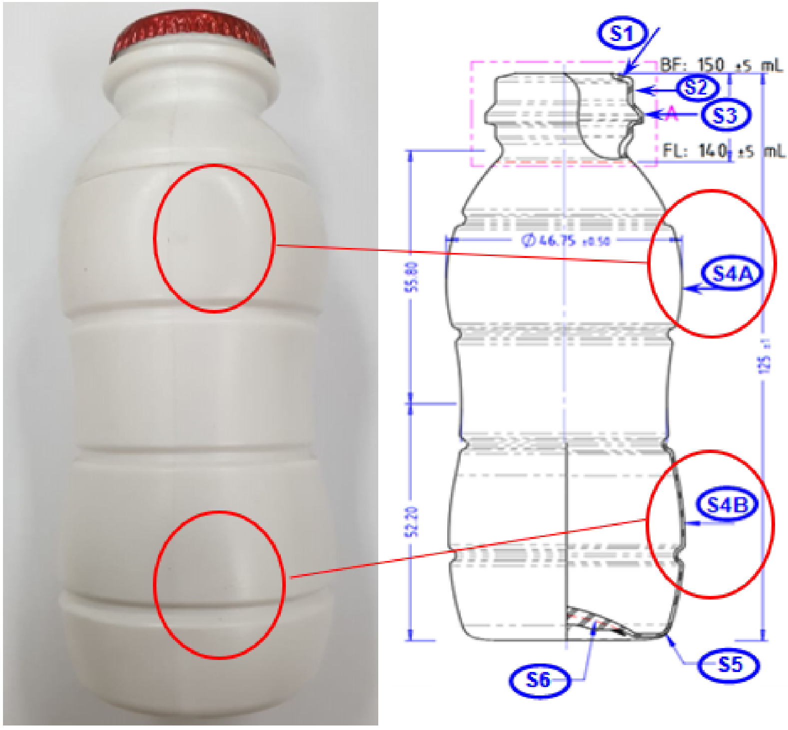

Figure 3: Position of dents in a defective bottle

Table 2: Physical properties of HDPE bottles (April 2019)

Parameters	Result	Standard	Unit	Note
Glass Transition	132.25	> 130	^{o}C	Pass
Wall thickness (S1)	1.1	0.9-1.2	mm	Pass
Wall thickness (S2)	1.1	0.8-1.2	mm	Pass
Wall thickness (S3)	0.4	0.5-1	mm	Pass
Wall thickness (S4A)	0.6	0.35-0.8	mm	Pass
Wall thickness (S4B)	0.6	0.35-0.8	mm	Pass
Wall thickness (S5)	0.6	0.35-0.8	mm	Pass
Wall thickness (S6)	1.3	0.6-1.6	mm	Pass
Sealing area	3.4	2.5-3.5	mm	Pass
O_2 Transfer	10.5	< 11.0	$\times$ 1010 cm^3/ cm^2/mm/sec/cmHg	Pass
H_2O Transfer	30.5	< 32.0	$\times$ 1010 cm^3/ cm^2/mm/sec/cmHg	Pass

Table 3: Cp and Cpk for bottle wall thickness parameters (S1-S6)

Sampling point	Cp	Cpk
S1	0.56	0.56
S2	0.57	0.57
S3	0.54	0.54
S4A	0.55	0.55
S4B	0.55	0.54
S5	0.56	0.57
S6	0.53	0.54

on the results shown in Table 2, it can be concluded that all the specified parameters were within the required standards. Release analysis is normally carried out for every bottle arrival once a year. The last release analysis was in April 2019. Plastic bottle thickness distribution and glass transition temperature are two possible packaging factors to cause dented bottles. Position of dents in a defective bottle after sterilization is shown in Figure 3. The thickness distribution is checked for every release and the glass transition temperature is checked annualy. The bottle thickness distribution, analyzed by Minitab 19, is shown in Table 3 and the bottles meet the specifications since Cp is equal to Cpk, as also indicated in Figure 4 and Figure 5. Cp is an index of short-term process capability, where calculations only pay attention to the distribution of data but do not pay attention to data centering. Cpk is an index of short-term process capability, where calculations focus on the distribution and centering of data (Aslam, Wu, Azam & Jun, 2013) It can be concluded that the material factor has no effect on a dented bottle.

The glass transition temperature of HDPE is 132.25 ^{o}C which is much higher than the sterilization temperature of 121.0 ^{o}C. Therefore, the sterilization temperature is unlikely to cause dented bottles. Other possible factors that contribute to dented bottles are the packaging permeability and the structure of the bottle. Permeability of HDPE to O_2 is 10.5 $\times$ 10^{10} cm^3/ cm^2/mm/sec/cmHg and to H_2O is 30.5$\times$ 1010 cm^3/ cm^2/mm/sec/cmHg. These permeability values meet the standards as specified in Table 2.

Ring design contributes to strength of the bottle structure. The HDPE bottles used in this study have 4 circular rings, with a diameter of 2 mm. The aluminium lid to seal the HDPE bottle has three different layers, i.e. aluminum, adhesive and LDPE. Table 3 indicates that all the lid parameters assessed are within the required standards and may not be the major cause of the dented bottle.

Method

Commercially sterilized milk is processed using a rotary over pressure retort. Process parameters which are important and may contribute to dented bottles are temperature, pressure and the percentage of the retort basket area occupied by bottles (bottle density). The temperature and pressure profiles during the sterilization process are shown in Figure 6. The maximum temperature experienced by products and the packaging material is 121.0 ^{o}C which is much lower than the glass transition temperature as already discussed above. More importantly, a process parameter that potentially causes dents on the plastic bottle is pressure difference. Figure 6 shows that external pressure in the retort is 2.00 bar and the internal pressure in the bottle is 3.20 bar. An imbalance between external pressure and internal pressure may deform packaging, especially for packaging materials that cannot stand a high pressure difference. Hariyadi (2017) stated that there is excess pressure on over-pressure retorts which serves to keep the packaging from expanding too much and causing damage to the packaging. When the retort pressure is too low, the packaging expands because the internal pressure is higher than the external pressure. A pressure imbalance causes the product to expand and bottles to contact each other leading to dents. From focus group discussions it was agreed that pressure in the retort during the sterilization process is the main possible cause of dented HDPE bottles. A plan to reduce the pressure difference was developed for process improvement. Another possible cause of dented bottles from the method perspective is the percentage of the retort basket area occupied by bottles (bottle density). The bottle density used during normal production

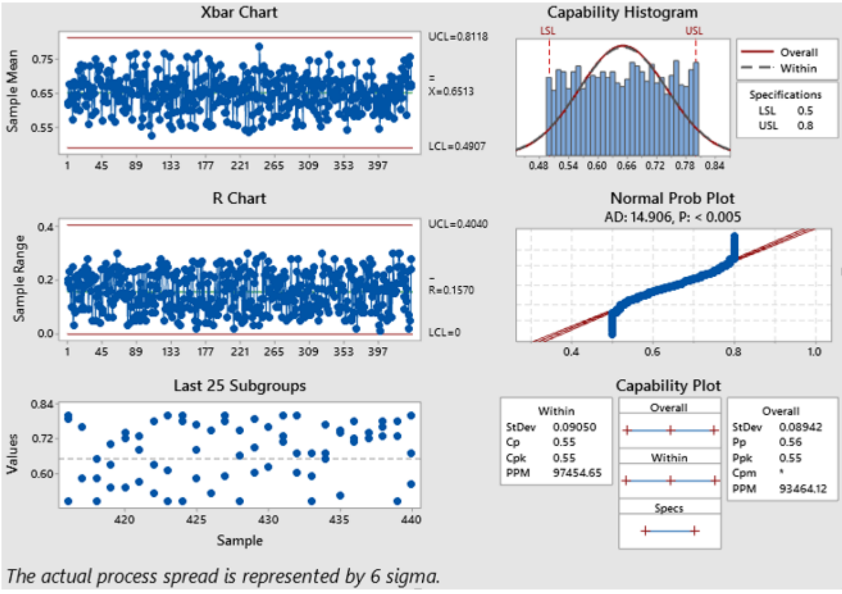

Figure 4: Control normal capability sixpack report on S4A for incoming bottles: March 2018-February 2019

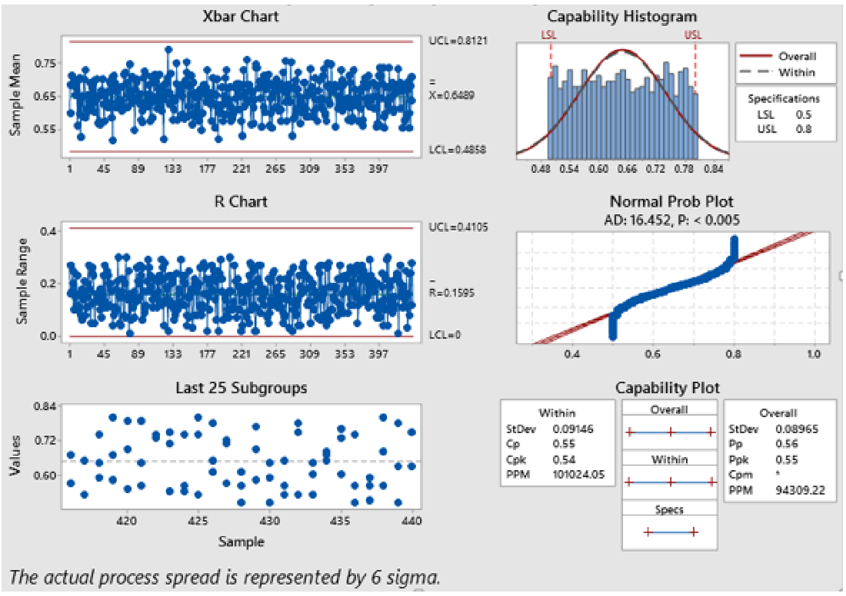

Figure 5: Control normal capability sixpack report on S4B for incoming bottles: March 2018-February 2019

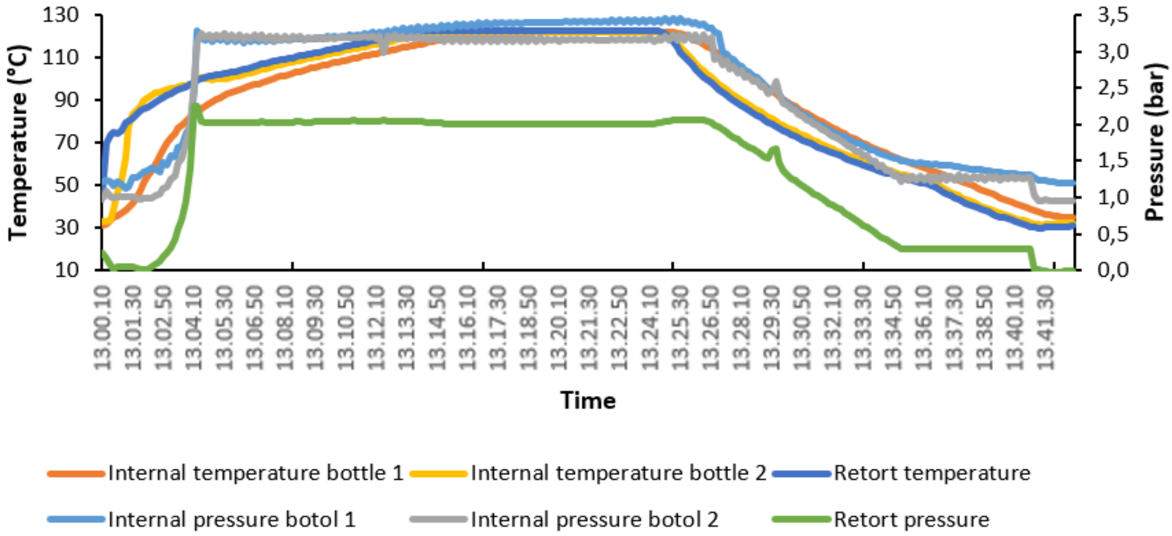

Figure 6: Chart of the sterilization process at retort sterilization temperature 121 ^{o}C, sterilization time 10 minutes, retort pressure 2.00 bar and 82% bottle density.

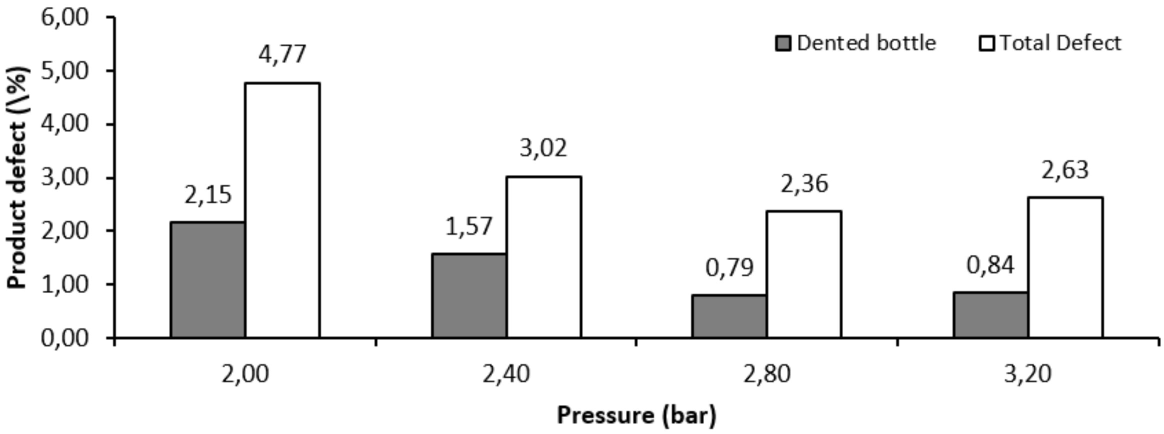

Figure 7: Percentage of product defects at various pressures, with retort sterilization temperature 121 ^{o}C, sterilization time 10 minutes and 82% bottle density.

Table 4: Physical properties of aluminium lids (April 2019)

Parameters	Result	Standard	Unit	Note
External limitation	Conform	Conform	-	Pass
Width Tolerance	116	114 - 118	mm	Pass
Thickness Tolerance	125	123 - 127	μm	Pass
Peel strength	10.28	> 3.5	N/15mm	Pass
Heat sealing	41.69	> 4 40.00	N/15mm	Pass
Sterilization at 121 ^{o}C (30 min)	Conform	Conform	-	Pass

routine is 82%. For futher improvement, experiments were carried out by reducing the bottle density from 82% to 77% and 73% at optimum retort pressure.

3.3 Improvement of the Largest Defect Category

The improvement plan to reduce dented bottle defects was executed by increasing retort pressure from 2.00 bar to 2.40 bar, 2.80 bar and 3.20 bar. The experimental results are shown in Figure 7, where an increasing pressure decreased the percentage of dented bottle. A better balance between internal and external pressure in the bottle reduces the number of dented bottles. Least dented bottles was obtained at a retort pressure of 2.80 bar, where 2.36% of all products produced were defective and 0.79% of all products were dented bottle defects. Based on this result, the retort pressure used in studying the effect of bottle density was 2.80 bar.

Reducing the number of defective HDPE bottles by increasing the retort pressure may affect the sterility level received by the products. Therefore, it was necessary to measure the sterility level (F_0 value) by performing a heat penetration test using 12 data loggers. F_0 values were calculated using a trapezoidal method, with a z value of 10 ^{o}C based on *Clostridium botulinum* (Membre & van Zuijlen, 2010). Minimum F_0 and maximum F_0 results, and their difference, are shown in Figure 8. All F_0 values were greater than 10 minutes which indicates that the sterilization process was sufficient.

The bottle density is the percentage of the retort basket area (6,496 cm2) occupied by bottles. A reduction in bottle density from 82% to 77% and 73% was expected to provide the right distance for bottles to expand. Based on the results shown in Figure 9, it can be concluded that reducing bottle density reduces total defects and dented bottles. However, there was an increase in striped lid defects caused by friction between the aluminum lid and spacer mat due to loose space when the basket rotates, and a reduction in productivity. Therefore, this strategy to reduce bottle density was considered ineffective as a solution for reducing product defects.

3.4 Verification of Improvement Results

According to Giwa et al. (2019), experiment results obtained on a process over a short-term period of time need to be verified over a long-term period of time. Studying the process over a long-term p period of time can accommodate other factors that did not appear during the short-term study of the process. The results of the long-term study over a three-month period, where 3,495,726 bottles were produced, are shown in Figure 10. 2.24% of all products sterlized were defective and 0.76% of all products were dented bottle defects. These results are comparable with those obtained during the production trial at 2.80 bar (2.63% of all products defective and 0.79% dented bottle defects). Therefore, it can be concluded that an increased retort pressure to maintain pressure balance in the HDPE bottles can result in a reduced number of dented bottle defects during the company's routine production. By applying a retort pressure of 2.80 bars can reduce the percentage of

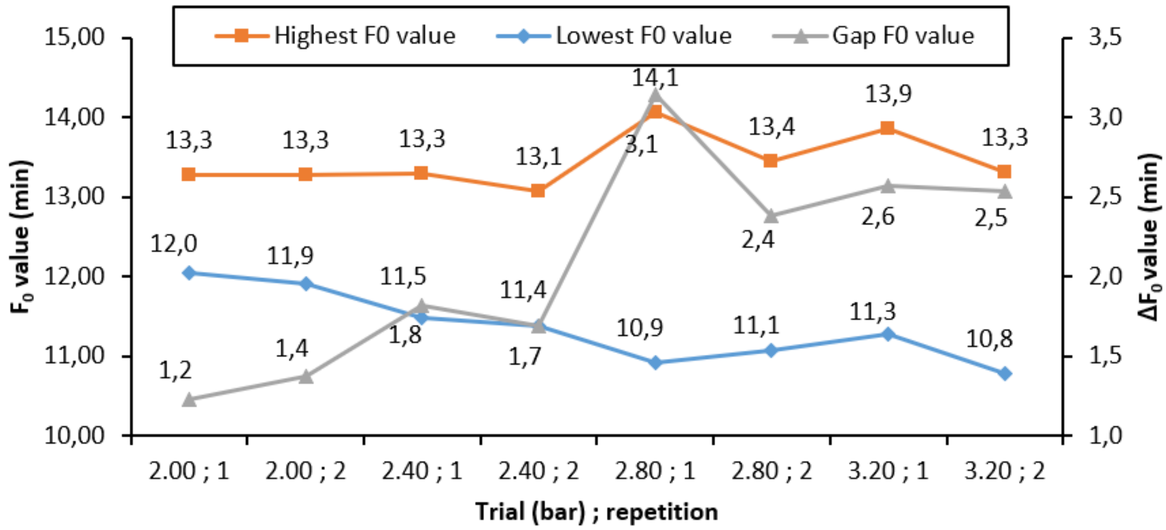

Figure 8: Sterilizing values (F_0) at various pressures, with retort sterilization temperature 121 ^{o}C, sterilization time 10 minutes, and 82% bottle density.

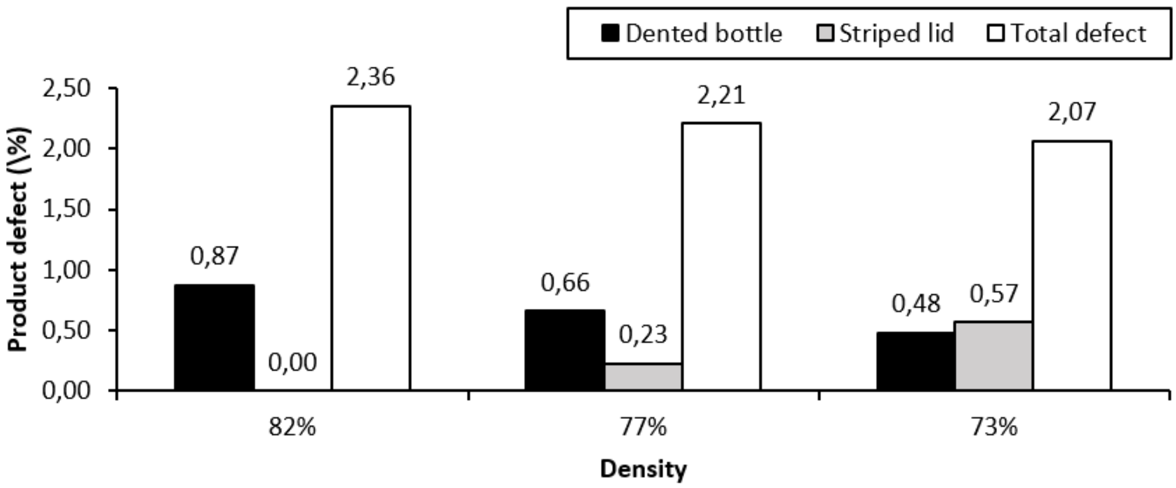

Figure 9: Percentage of product defects at various bottle densities, with retort pressure 2.80 bar, sterilization temperature 121 ^{o}C and sterilization time 10 minutes.

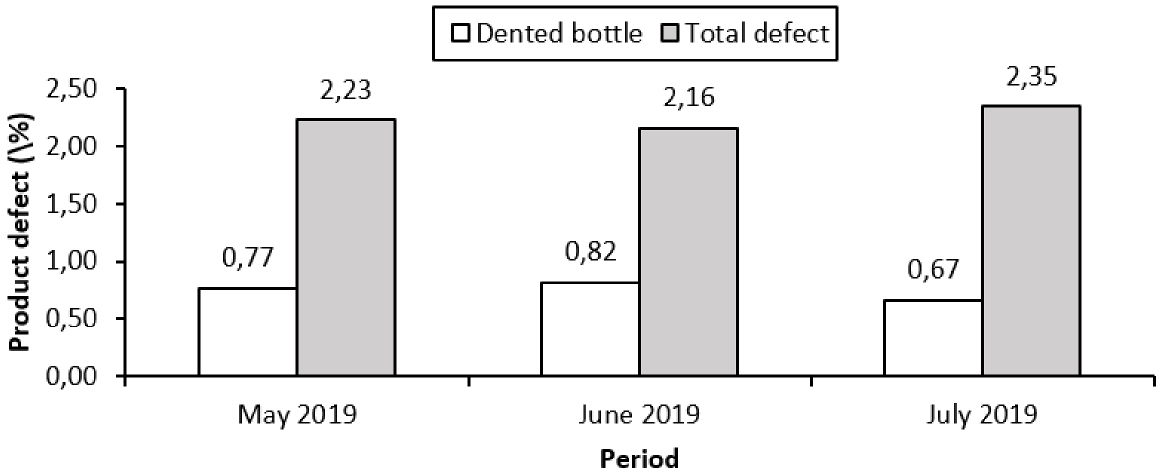

Figure 10: Percentage of product defects during three-month production, with retort pressure 2.80 bar, sterilization temperature 121 °C, sterilization time 10 minutes and 82% bottle density.

defective products produced by the company by 2.9%. Based on the monthly production volume, the company gains an additional output of 73,080 bottles which is equivalent to USD 16,733 per month.

4 Conclusion

Production of commercially sterilized millk in HDPE bottles experienced product loss, where 5.14% of all products produced were defective and 2.37% of all products were dented bottle defects. Dented HDPE bottles was the largest defect category based on Pareto analysis. Based on the cause-effect diagram, the root cause of the dented bottles was a pressure imbalance experienced by the bottles during the sterilization process. The retort pressure was 2.00 bar, while the internal pressure was 3.20 bar. These conditions cause the product to expand and bottles to contact each other leading to dents. Production trials carried out at a retort pressure of 2.80 bar improved the process, where 2.36% of all products produced were defective and 0.79% of all products were dented bottle defects. Applying a lower bottle density during the sterilization process could decrease the number of dented bottles, however, it increased the number of striped lid defects and reduced productivity. The best conditions for sterilization (retort pressure = 2.80 bar; number of bottles/baskets = 1938 bottles) were verified over a three-month full-scale production trial. By applying a retort pressure at 2.80 bar the percentage of defective products could be reduced by 2.9%, and in return this could save the company up to 73,080 bottles monthly (equivalent to USD 16,733).

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Functional and Pasting Properties of *Gari* Produced from White-fleshed Cassava Roots as Affected by Packaging Materials and Storage Periods, and Sensory Attributes of the Cooked *Gari* Dough (*eba*)

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Abstract

Gari (roasted fermented cassava grits) is the most popular product consumed in West Africa and an important food product in the diet of millions of people in developing countries. The study investigated the effect of packaging materials (PM) and storage periods on the functional and pasting properties of *Gari* produced from white-fleshed cassava roots and sensory attributes of the cooked *Gari* dough (*eba*).*Gari* was produced using the standard method and packaged in a polypropylene woven sack (PP) and polyvinyl chloride container (PVC). *Gari* was stored for 24 weeks at room temperature and sampled at four-week intervals for functional and pasting properties, and sensory evaluation of the *eba* , using standard methods. The results showed that the storage periods significantly affected all the functional (except swelling power) and pasting properties of the *Gari*, and PM had no significant ($p>0.05$) effect on the functional (except bulk density) and pasting properties. Also, the PM had no significant effect on the sensory attributes of the *eba* except for the mouldability ($p<0.05$). The setback viscosity of the *Gari* packaged in PVC had a significant ($p<0.05$, $r= -0.58$) negative correlation with the texture of the *eba*. The panellists preferred all the sensory attributes of the *eba* made from the *Gari* stored in PP compared to that made from *Gari* stored in PVC. Therefore, packaging *Gari* in PP may keep most of the properties preferred by consumers when stored for up to 5 months.

Keywords: *Gari*; Packaging materials; Storage period; Sensory evaluation; *eba*

1 Introduction

Cassava (*Manihot esculenta*) is the third-largest source of carbohydrates in the tropics, after rice and maize (Fauquet & Fargette, 1990). Cassava root is a primary staple food in the developing world, providing an essential diet for over half a billion people (FAO, 1995). Nigeria is the largest producer of cassava root in the world, whilst Thailand is the largest exporter of cassava products. At present, cassava is undergoing a transition from a mere subsistence crop found in the fields of peasants to a commercial plant grown in plantations. This expansion is attributed to its discovery as a major source of food carbohydrate that could be processed into different forms of human delicacies and animal feeds (Simonyan et al., 2014).

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Cassava has been a source of raw materials for many industrial products such as starch, flour, and ethanol. Cassava production is relatively easy as it is tolerant of the biotic and edaphic factors that hamper the production of other crops. Cassava roots are used to store energy, unlike the roots of sweet potato and yam tuber that are reproductive organs. Despite their agronomic advantages, root crops are far more perishable than other staple food crops. Westby (2002) reported that cassava has a shelf life that is generally accepted to be of the order of 24 to 48 h after harvest.

Cassava utilization patterns vary considerably in different parts of the world, and in Nigeria, the majority of the cassava produced (90%) is used for human food. Thus, cassava, in its processed form, is a reliable and convenient source of food for ten million rural and urban dwellers in Nigeria (IITA, 2010). However, fresh cassava has a limited storage life because of its high moisture content, and its processing into relatively shelf-stable intermediate and final products for various food applications is therefore necessary (Quaye, Gayin, Yawson & Plahar, 2009). Cassava can be transformed into different products such as *Gari, fufu, lafun,* and many other West African traditional dishes (Afoakwa, Kongor, Annor & Adjonu, 2010).

Gari is a roasted, fermented cassava meal, and the most important food product in the diet of millions of Nigerians and Ghanaians (Afoakwa et al., 2010). *Gari* is a staple food that used to be within the purchasing power of many people in the society irrespective of their income and status (Sanni, Adebowale, Awoyale & Fetuga, 2008). *Gari* is produced by grating fresh roots into a mash, fermenting, de-watering, granulating, sifting, and roasting into gelatinized particles (James et al., 2012). However, there is a need to package the *Gari* before marketing and subsequent storage.

The storage stability of food systems depends on the storage conditions, packaging materials, and the water activity of the food material (Ilouno, Ndimele, Adikwu & Obiekezie, 2016; Okigbo, 2003). The deterioration of floury products is usually attributed to the type of packaging materials and spoilage organisms, such as bacteria and fungi. Various studies have examined the effect of different packaging materials (high and low-density polyethylene bags, polypropylene woven sacks and containers) and storage conditions on the quality attributes of *Gari* (Adebowale et al., 2017; Adejumo & Raji, 2012; FAO, 1999; Ogiehor & Ikenebomeh, 2006). However, at the time of this study, to the best of our knowledge, no work has been reported on the sensory acceptability of cooked paste (*eba*) from *Gari* as it relates to the effect of different packaging materials and storage conditions. Hence, this study aims to evaluate the impact of packaging materials and storage periods on the functional and pasting properties of *Gari* produced from white-fleshed cassava roots, and sensory attributes of the cooked paste (*eba*).

2 Materials and Methods

2.1 Materials

Freshly harvested cassava roots [tropical Manioc esculenta 419 (TME419)] were obtained from the farms of IITA Ibadan. One hundred and fifty kilograms (150 kg) of the cassava roots were then processed into *Gari.* The packaging materials (PVC and PP) were obtained from a local market (Aleshinolye) in Ibadan, Oyo State, Nigeria.

2.2 Processing of Cassava roots into *Gari*

Gari was produced using the method described by Abass, Dziedzoave, Alenkhe and James (2013). Healthy cassava roots were peeled, washed, and grated. The resulting mash was packed in a polypropylene (PP) woven sack, which was then put in a fermenter. The grated cassava mash was allowed to ferment for 72 h under ambient temperature. The fermented mash was dewatered and sieved to remove fibrous materials and then roasted. The *Gari* was allowed to cool and packed in polyethylene bags for further study

2.3 Storage studies of *Gari* samples

Five hundred grams of the *Gari* produced were weighed and packaged in a PP woven sack, sealed, and placed in a PVC container and covered with a close-fitting lid. This was stored at room temperature (28 - 30 ^oC) for 24 weeks inside a cupboard. The PP packaging material had a thickness of 0.75 μm, oxygen permeability of 160 mm/100 cm^2 in 24 h and 25 ^oC) and water vapour permeability of 0.27 g/100 cm^2 in 24h at 37.8 ^oC and relative humidity of 90%. The PVC packaging material had a thickness of 0.45 μm, oxygen permeability of 80 mm/100 cm^2 in 24 h and 25 ^oC) and water vapour permeability of 8 g/100 cm^2 in 24h, 37.8^oC and relative humidity of 90%, as reported by Awoyale, Maziya-Dixon and Menkir (2016). The functional and pasting properties of the *Gari* and sensory attributes of the cooked paste (*eba*) were determined every 4 weeks of the 24 weeks storage periods as described by Abiodun, Adegbite and Oladipo (2010).

2.4 Determination of functional properties of *Gari* samples

Bulk Density

Bulk density was determined using the methods described by Ashraf, Anjum, Nadeem and Riaz (2012). *Gari* sample (10 g) was measured into a graduated measuring cylinder (50 ml) and lightly tapped on the workbench (10 times) to attain a constant height. The bulk density was then recorded and expressed as grams per millilitre.

Determination of swelling power and solubility index

For the determination of swelling power and solubility index, aqueous starch dispersions of 2.5% were put in centrifuge tubes, capped to prevent spillage, and heated in a water bath with shaker (Precision Scientific, Model 25: Chicago, USA) at 85^oC for 30 min. (Afoakwa & Nyirenda, 2012). The samples were cooled to room temperature and centrifuged (Thelco GLC- 1, 60647: Chicago, USA) at 3,000 rpm for 15 min. The weight of the precipitated paste separated from the supernatant was taken, after which a hot air oven (Memmert GmbH+Co.KG: D-91126, Germany) was used to evaporate the supernatant at 105 ^oC, and the residue weighed. The swelling power (SWP), and solubility index (SI) were then calculated as shown in equation 1 and 2:

$$\text{SWP} = \frac{\text{Wt of precipitated paste}}{\text{Wt of sample}} - \text{Wt of residue in the supernatant} \tag{1}$$

$$\text{SI} = \frac{\text{Wt of residue in the supernatant}}{\text{Wt of sample}} \times 100 \tag{2}$$

Water absorption capacity

The water absorption capacity (WAC) of *Gari* was determined as described by Oyeyinka et al. (2013) with a few modifications. *A 1g* sample of *Gari* was weighed into a clean pre-weighed dried centrifuge tube and mixed thoroughly with 10 ml distilled water by vortexing after which the suspension was allowed to stand for 30 min and centrifuged (Thelco GLC- 1, 60647: Chicago, USA) at 3,500 rpm for 30 min. The supernatant was decanted after centrifugation, with the tube and the sediment weighed. The weight of water (g) retained in the sample was reported as WAC.

Oil absorption capacity

A sample of *Gari* (1 g) was suspended in 5 ml of vegetable oil in a centrifuge tube, after which the slurry was shaken on a platform tube rocker for 1 min at ambient temperature and centrifuged (Thelco GLC- 1, 60647: Chicago, USA) at 3000 rpm for 10 min. The supernatant was decanted and discarded. The adhering drops of oil were removed and reweighed. The oil absorption capacity (OAC) was expressed as the weight of the sediment/initial weight of the sample (g/g) (Awoyale et al., 2020).

$$Dispersibility(\%) = \frac{(50 - volumeofthesettleparticule)}{50} \times 100 \tag{3}$$

Dispersibility

The pasting properties of the *Gari* samples were measured using a Rapid Visco Analyser

(Model RVA 4500, Perten Instruments, Australia) equipped with a 1000 cmg sensitivity cartridge. *Gari* (3.5 g) was weighed into a dried empty canister, after which 25 ml of distilled water was added to the sample. The mixture was stirred as prescribed, and the canister fitted into the RVA as recommended. The mixture was then heated from 50 to 95 ^{o}C at a rate of 1.5 ^{o}C/min, over a period of 15 min inside the Visco Analyzer, and cooled to 50 ^{o}C. The viscosity profile indices that were recorded from the pasting profile with the aid of Thermocline for Windows Software connected to a computer were peak viscosity, trough, breakdown, final viscosity setback, peak time, and pasting temperature (Falade & Olugbuyi, 2010).

2.5 Preparation of *eba* for sensory evaluation

The *Gari* was cooked into a dough (*eba*) using the modified method described by Udoro, Kehinde, Olasunkanmi, Charles et al. (2014). The *eba* was prepared by adding about 100 g of *Gari* to 195 ml of boiling water (100 ^{o}C) and continuously stirred to form a smooth thick paste. The sensory evaluation was carried out using twelve trained panellists from the staff and graduate students of the International Institute of Tropical Agriculture (IITA), Ibadan who consume *eba* regularly, on the attibutes colour/appearance, texture, stretchability, mouldability, flavour, mouthfeel, and overall acceptability. The sensory acceptability of the *eba* produced from *Gari* before and after each of the storage periods (4 weeks) for 24 weeks was evaluated using a 9-point hedonic scale as reported by Iwe (2002). The authors of this study declare that, the sensory evaluation followed the tenets of the Declaration of Helsinki promulgated in 1964 and was approved by the institutional ethical review committee. In addition, verbal consent was obtained from the participants.

2.6 Statistical analysis

Analysis of variance (ANOVA), separation of the mean values (using Duncan's Multiple Range Test at $P < 0.05$) and the Pearson correlation were calculated using the Statistical Package for Social Scientists (SPSS) software (SPSS Inc., Chicago, IL version 21.0) (Awoyale, Sanni, Shittu & Adegunwa, 2015; Awoyale et al., 2020).

3 Results and Discussion

3.1 Effect of storage periods and packaging materials on the functional properties of stored *Gari* samples

The functional properties of a material determine its application and end-use (Adeleke, Odedeji et al., 2010). That is, the functional properties indicate how the food materials under examination will interact with other food components directly or indirectly affecting the processing applications, food quality, and ultimate acceptance. The effects of storage periods and packaging materials on the functional properties of the *Gari* are presented in Table 1. The WAC represents the ability of a product to associate with water under conditions where water is limited, and it was observed to be significantly ($p<0.001$) affected by storage periods. WAC of the *Gari* sample ranged from 445.54% to 524.95% in PP, and 445.54% to 533.76% in PVC, which were within the range reported by Awoyale et al. (2016) and Olanrewaju and Idowu (2017), this variation may be attributed to different storage periods and differences in the granule structure, and degrees of availability of the water binding sites among the samples (Xian, Shariffa & Azwan, 2020). No combined interactive effects ($p>0.05$) (Table 1) of storage periods and packaging materials were observed on the functional properties except for bulk density (BD) and solubility index (SI) ($p<0.001$). However, a significant and positive correlation ($p<0.01$; $r = 0.56$) existed between the WAC for stored *Gari* samples and the overall acceptability of panellists for *eba*, and the same result was observed for mouldability ($p<0.05$; $r = 0.51$) and mouthfeel ($p<0.01$; $r = 0.46$), respectively (Table 1).
The OAC is a measure of the ability of food material to absorb oil, which acts as a flavour retainer and improves the mouthfeel of foods generally (Awuchi, Igwe & Echeta, 2019). A sig-

Table 1: Effect of storage period and packaging materials on the functional properties of *gari*

Parameters	Storage wks	*Gari* stored in PP Mean	*Gari* stored in PP Range	*Gari* stored in PVC Mean	*Gari* stored in PVC Range	p value of Package	p value of Storage periods	p value of Storage period x Package
WAC (%)	0	445.54±3.25^{a}	443.24 – 448.84	445.54±3.25^{a}	443.24 – 447.84			
	4	456.75±0.57^{a}	456.35 – 457.15	533.76±4.59^{c}	499.40 – 568.12	NS	***	NS
	8	524.95±2.14^{b}	523.43 – 526.46	509.57±3.22^{bc}	507.29 – 511.84	NS	***	NS
	12	495.58±3.12^{b}	493.37 – 497.78	514.87±9.98^{bc}	507.81 – 521.93	NS	***	NS
	16	442.53±2.93^{a}	440.28 – 444.60	452.78±8.03^{a}	447.11 - 458.45	NS	***	NS
	20	500.71±3.14^{b}	477.28 – 524.14	476.88±14.50^{ab}	466.63 – 568.12	NS	***	NS
OAC (%)	0	140.10±6.57^{a}	135.45 – 144.74	140.10±6.57^{cd}	135.45 – 144.74			
	4	126.'17±1.45^{a}	118.08 – 134.27	115.91±7.05^{a}	110.64 – 120.62	NS	***	NS
	8	139.61±6.51^{a}	135.00 – 144.21	152.91±1.44^{d}	151.90 – 153.93	NS	***	NS
	12	135.77±7.40^{a}	130.54 – 141.00	136.40±8.13^{c}	130.65 – 142.15	NS	***	NS
	16	141.68±4.63^{a}	138.40 – 144.96	132.81±4.50^{bc}	129.63 – 135.99	NS	***	NS
	20	121.70±1.93^{a}	110.43 – 132.96	121.07±2.61^{ab}	119.23 – 122.91	NS	***	NS
SP (%)	0	11.54±0.26^{a}	11.36 – 11.72	11.54±0.26^{a}	11.36 – 11.72			
	4	11.77±0.05^{a}	11.73 – 11.80	12.15±0.40^{a}	11.86 – 12.43	NS	NS	NS
	8	12.31±0.79^{a}	11.76 – 12.87	12.06±1.71^{a}	10.85 – 13.27	NS	NS	NS
	12	10.74±0.53^{a}	10.36 – 11.11	11.26±0.60^{a}	10.83 – 11.69	NS	NS	NS
	16	14.84±1.13^{a}	14.04 – 15.64	13.32±1.20^{a}	12.47 – 14.17	NS	NS	NS
	20	11.70±0.01^{a}	11.69 – 11.71	12.19±0.88^{a}	10.83 – 14.17	NS	NS	NS
SI (%)	0	37.98±1.06^{b}	37.23 – 38.73	37.98±1.06^{bc}	37.23 – 38.73			
	4	37.38±7.04^{b}	32.41 – 42.36	35.14±2.56^{b}	33.32 – 36.96	NS	***	NS
	8	38.06±3.15^{b}	35.83 – 40.28	39.12±1.21^{bc}	38.26 – 39.95	NS	***	NS
	12	45.82±2.92^{b}	43.75 – 47.88	44.32±4.2^{c}	41.34 – 47.30	NS	***	NS
	16	14.00±5.59^{a}	10.05 – 17.96	15.12±3.52^{a}	12.63 – 17.61	NS	***	NS
	20	39.41±7.80^{b}	11.69 - 15.64	43.10±4.91^{bc}	39.62 – 46.57	NS	***	NS
BD (g/ml)	0	0.57±0.01^{b}	0.57 – 0.58	0.57±0.01^{bc}	0.57 – 0.58			
	4	0.57±0.00^{b}	0.57 – 0.57	0.61±0.11^{c}	0.60 – 0.62	***	***	***
	8	0.59±0.03^{b}	0.57 – 0.62	0.55±0.03^{ab}	0.53 – 0.57	***	***	***
	12	0.55±0.03^{b}	0.53 – 0.57	0.51±0.02^{a}	0.50 – 0.53	***	***	***
	16	0.56±0.01^{b}	0.56 – 0.57	0.57±0.02^{bc}	0.56 – 0.59	***	***	***
	20	0.50±0.00^{a}	0.50 – 0.50	0.57±0.00^{bc}	0.57 – 0.57	***	***	***
Dispersibility (%)	0	45.50±0.71^{a}	45.00 – 46.00	45.50±0.71^{a}	45.00 – 46.00			
	4	46.25±0.35^{a}	46.00 – 46.50	45.75±0.35^{a}	45.50 – 46.00	NS	***	***
	8	59.00±0.00^{c}	59.00 – 59.00	53.00±0.00^{b}	53.00 – 53.00	NS	***	***
	12	44.00±0.00^{a}	44.00 – 44.00	56.00±0.00^{c}	56.00 – 56.00	NS	***	***
	16	44.50±0.71^{a}	44.00 – 45.00	47.00±1.41^{a}	46.00 – 48.00	NS	***	***
	20	49.50±2.12^{b}	48.00 – 51.00	51.00±1.41^{b}	50.00 – 52.00	NS	***	***

WAC: Water absorption index, OAC: Oil absorption capacity, SP: solubility power, SI: Solubility index, BD: Bulk density, PP: Polypropylene woven sack, PVC: Polyvinyl chloride container. NS: not significant ($p>0.05$); *$p<0.05$, **$p<0.01$, ***$p<0.001$, Means in the same row and followed by the same letters are not significantly different from each other ($p>0.05$)

Table 2: Effect of storage period and packaging materials on the pasting properties of *gari*

Parameters	Storage wks	*Gari* stored in PP Mean	*Gari* stored in PP Range	*Gari* stored in PVC Mean	*Gari* stored in PVC Range	p value of Package	p value of Storage periods	p value of Storage period x Package
Peak viscosity (RVU)	0	653.63 ± 34.71^{d}	629.08-678.17	653.63 ± 34.71^{d}	629.08-678.17			
	4	494.96 ± 22.80^{c}	478.83-511.08	415.84 ± 8.25^{c}	410.00-421.67	NS	***	*
	8	372.46 ± 18.33^{b}	359.50-385.42	347.75 ± 4.95^{ab}	344.25-351.25	NS	***	*
	12	352.75 ± 2.23^{b}	351.17-354.33	353.96 ± 15.73^{b}	342.83-365.08	NS	***	*
	16	276.75 ± 24.40^{a}	259.50-294.00	326.13 ± 14.91^{ab}	315.58-336.67	NS	***	*
	20	276.46 ± 8.90^{a}	270.17-282.75	308.83 ± 6.01^{a}	304.58-313.08	NS	***	*
	0	383.83 ± 17.32^{c}	371.58-396.08	383.83 ± 17.32^{d}	371.58-396.08			
	4	354.92 ± 13.31^{c}	3.50-364.33	317.63 ± 5.37^{c}	313.83-321.42	NS	***	**
	8	280.21 ± 9.84^{b}	273.25-287.17	267.29 ± 4.65^{b}	264.00-270.58	NS	***	**
	12	272.75 ± 11.78^{b}	264.42-281.08	261.13 ± 35.18^{ab}	236.25-286.00	NS	***	**
	16	245.2 ± 34.83^{ab}	220.58-269.83	286.80 ± 16.79^{bc}	274.92-298.67	NS	***	**
	20	210.50 ± 7.54^{a}	205.17.215.83	221.21 ± 3.13^{a}	219.00-223.42	NS	***	**
Breakdown viscosity (RVU)	0	269.79 ± 17.38^{d}	257.17-282.08	269.79 ± 17.38^{c}	257.50-282.08			
	4	140.04 ± 9.49^{c}	133.33-146.75	98.21 ± 13.61^{b}	88.58-107.83	NS	***	NS
	8	92.25 ± 8.49^{b}	86.25-98.25	80.46 ± 0.30^{b}	80.25-80.67	NS	***	NS
	12	80.00 ± 9.55^{b}	73.25-86.75	92.83 ± 19.45^{b}	79.08-106.58	NS	***	NS
	16	31.55 ± 10.43^{a}	24.17-38.92	39.34 ± 1.89^{a}	38.00-40.67	NS	***	NS
	20	65.96 ± 1.36^{b}	65.00-66.92	87.63 ± 2.89^{b}	85.58-89.67	NS	***	NS
	0	568.96c$\pm9.37^{c}$	562.33-575.58	568.96 ± 9.37^{d}	179.50-190.75			
	4	503.38 ± 15.49^{b}	492.42-514.33	456.84 ± 2.60^{c}	137.25-141.17	NS	**	NS
	8	405.75 ± 4.13^{a}	402.83-408.67	400.13 ± 5.95^{a}	131.92-133.75	NS	**	NS
	12	423.84 ± 10.73^{a}	416.25-431.42	424.34 ± 3.77^{b}	135.67-190.75	NS	**	NS
	16	415.04 ± 3.24^{a}	412.75-417.33	434.29 ± 10.66^{b}	143.17-151.83	NS	**	NS
	20	401.46 ± 2.06^{a}	400.00-402.92	415.84 ± 13.91^{ab}	187.00-202.25	NS	**	NS
Setback viscosity (RVU)	0	185.13 ± 7.96^{b}	179.50-190.75	185.13±5.63bc	5.00-5.20			
	4	148.46 ± 2.18^{ab}	146.92-150.00	139.21 ± 1.96^{a}	5.20-5.27	NS	*	NS
	8	125.54 ± 5.71^{a}	121.50-129.58	132.84 ± 0.91^{a}	5.40-5.53	NS	*	NS
	12	151.09 ± 22.51^{ab}	135.17-167.00	163.21 ± 27.54^{abc}	5.33-5.87	NS	*	NS
	16	169.84 ± 31.59^{b}	147.50-192.17	147.50 ± 6.12^{ab}	5.93-6.07	NS	*	NS
	20	190.96 ± 26.57^{b}	187.08-194.83	194.63 ± 27.09^{c}	5.20-5.47	NS	*	NS
Peak time (Min)	0	5.10 ± 0.14^{a}	5.00-5.20	5.10 ± 0.10^{a}	5.00-5.20			
	4	5.17 ± 0.05^{ab}	5.13-5.20	5.24 ± 0.04^{a}	5.20-5.27	NS	***	NS
	8	5.34 ± 0.09^{ab}	5.27-5.40	5.47 ± 0.07^{a}	5.40-5.53	NS	***	NS
	12	5.60 ± 0.00^{ab}	5.60-5.60	5.60 ± 0.27^{ab}	5.33-5.87	NS	***	NS
	16	6.23 ± 0.42^{c}	5.93-6.53	6.00 ± 0.07^{b}	5.93-6.07	NS	***	NS
	20	5.37 ± 0.05^{bc}	5.33-5.40	5.34 ± 0.14^{a}	5.20-5.47	NS	***	NS
Pasting temperature (^{o}C)	0	80.70 ± 0.07^{a}	80.65-80.75	80.70±0.05a	80.65-80.75			
	4	80.73 ± 0.04^{a}	80.70-80.75	81.50 ± 0.05^{a}	81.45-81.55	NS	***	NS
	8	82.35 ± 1.06^{ab}	81.60-83.10	83.28 ± 0.03^{ab}	83.25-83.30	NS	***	NS
	12	83.13 ± 0.04^{ab}	83.10-83.15	83.18 ± 0.03^{ab}	83.15-83.20	NS	***	NS
	16	91.55 ± 5.09^{c}	87.95-95.15	87.23 ± 3.23^{b}	84.00-90.45	NS	***	NS
	20	87.13 ± 1.10^{bc}	86.35-87.90	83.23 ± 0.08^{ab}	83.15-83.30	NS	***	NS

PP: Polypropylene woven sack, PVC: Polyvinyl chloride container. NS: not significant (p>0.05); *p<0.05, **p<0.01, ***p<0.001, Means in the same row and followed by the same letters are not significantly different from each other (p>0.05)) nificant difference ($p<0.05$) was observed in the OAC of *Gari* stored in PVC, with no significant difference ($p>0.05$) for that stored in PP. The OAC obtained for this study ranged between 121.70% to 141.68% in PP and 115.91 to 152.91% in PVC, which were similar to the values reported by Awoyale, Sanni, Shittu, Adebowale and Adegunwa (2019) (Table 1).

Swelling power (SP) of starchy foods reveals the extent of associative forces within the granules; thus, the higher the SP, the lower the associative forces (Sanni, 2005). Good quality *Gari* is one that can swell up to at least 3 times its original size (Awoyale et al., 2020). The results showed that *Gari* stored in PP with higher SP (14.84%) had granules with lower associative forces when compared with *Gari* stored in PVC with lower SP (13.32%) (Table 1). This study showed no significant difference ($p > 0.05$) in the SP of the *Gari* during storage periods. Similarly, there was no significant ($p>0.05$) effect of the packaging materials and the interactions between the storage period and the packaging material on the stored *Gari*. The Solubility index (SI), which is related to the extent of leaching of amylose out of starch granules during swelling and affected by intermolecular forces, and the presence of surfactants and other associated substances (Awoyale et al., 2020) of the stored *Gari* ranged from 14.00% to 45.82%. A significant difference ($p<0.05$) existed in the SI during the storage periods. The SI of *Gari* packaged in PP had a significant positive correlation ($p<0.05$, $r = 0.61$) with the overall acceptability of the *eba*, and a negative ($p<0.05$) correlation with peak time ($r = -0.62$) and pasting temperature ($r = - 0.63$) (Table 3). Similar correlations were observed with *Gari* packaged in PVC, but this was not significant ($p>0.05$) (Table 4).

The bulk density (BD) is critical to evaluate floury products with respect to the weight, handling requirements and the type of packaging materials suitable for storage and transportation of food materials, (David, Arthur, Kwadwo, Badu & Sakyi, 2015). Similarly, the BD of a product is an essential parameter in determining suitable packaging materials and materials handling during food processing (Adebowale, Adegoke, Sanni, Adegunwa & Fetuga, 2012). Here, the BD of the *Gari* varied from 0.50 - 0.61 g/ml which was desirable and fell within an acceptable range of 0.50 g/ml to 0.91 g/ml as reported by Adindu and Aprioku (2006), and also agreed with the values 0.52 - 0.62 g/ml indicated by Nwancho, Ekwu, Mgbebu, Njoku and Okoro (2014), and Awoyale et al. (2016) (0.57 g/ml for white *Gari* and 0.56 g/ml for yellow *Gari*). Either loose or packed BD is influenced by factors such as dryness and particle size distribution of samples; thus the values obtained in this study were comparable to those reported by Udoro et al. (2014) (0.50 - 0.65g/cm^3). Komolafe and Arawande (2010) reported that the lower the BD value, the higher the amount of *Gari* that could be packaged in each volume of the container, which decreases the space occupied, the packaging, and transportation costs. However, *Gari* samples stored in PVC were observed to have higher BD, which was attributed to the packaging material. The storage and packaging materials had a significant ($p<0.001$) effect on the BD of *Gari*. A similar, significant ($p<0.01$) trend was observed for the combined interactive effect of the storage period and packaging materials of the *Gari*. The BD of the *Gari* packaged in PP was positively correlated ($p<0.05$, $r = 0.58$) with the mouldability while a positive and non-significant correlation existed with the trough viscosity ($p>0.05$, $r= 0.54$). However, a negative and significant correlation existed between the BD of the *Gari* packaged in PVC ($p<0.01$, $r = -0.81$) and the dispersibility of the *Gari* (Table 4).

Dispersibility is a measure of the reconstitution of flour starch in water, the higher the dispersibility, the better samples reconstitute in water as reported by Adebowale, Sanni and Fadaunsi (2008) and Awoyale et al. (2020). Dispersibility varied from 44.00% to 59.00% in PP and 45.50% to 56.00% in PVC, which agreed with an earlier study reported by Awoyale, Abass, Ndavi, Maziya-Dixon and Sulyok (2017) (43.22) on *Gari* (Table 2). Similarly, *Gari* stored (59.00%) in PP showed a significantly higher dispersibility value than *Gari* stored in PVC. This should be easily reconstituted in water without lump formation due to high dispersibility, whilst lump formation may likely occur in the *Gari* stored in PVC (56.00%) when soaked in water because of its low dispersibility Awoyale et al. (2020). The packaging materials had no significant effect ($p>0.05$)

on the dispersibility of the stored *Gari*. The storage periods and the interaction between the packaging materials and the storage periods had a significant impact ($p<0.01$) on the stored *Gari*. However, the dispersibility of the *Gari* packaged in PP had no significant correlation ($p>0.05$) on the functional, pasting and sensory properties of the *eba*.The *Gari* packaged in PVC had a negative correlation ($p<0.05$) with the trough viscosity ($r = -0.64$), and the final viscosity ($r = -0.63$) (Table 4).

3.2 Effect of storage periods and packaging materials on the pasting properties of *Gari*

Table 2 shows the effect of storage periods and packaging materials on the pasting properties of *Gari*. The pasting properties of flour products are used in assessing the suitability of its application as a functional ingredient in food products (Oluwalana, Oluwamukomi et al., 2011). Thus, the pasting properties of food products are essential in predicting their behaviour during and after cooking. The results of this study showed that the packaging materials had no significant ($p>0.05$) effect on the pasting properties of the *Gari*, but the storage periods had a significant impact ($p<0.05$) on the pasting properties (Table 2).

The peak viscosity (PV) is the maximum viscosity developed during or soon after the heating of the floury product (Adebowale et al., 2008) and here it ranged from 276.46 RVU to 653.63 RVU, similar to the values reported by Nwancho et al. (2014) (322.67 RVU) for *Gari* produced from dried cassava chips. On the other hand, values obtained for this study were higher than values recorded by Awoyale et al. (2017), Awoyale et al. (2019) (241.30 RVU; 183 RVU). However, PV is often related to the final product quality, as it indicates the viscous load faced during mixing (Maziya-Dixon, Dixon & Adebowale, 2007). The PV of the *Gari* packaged in PP had a significant positive correlation with the trough viscosity ($p<0.01$, $r = 0.96$), breakdown viscosity ($p<0.01$, $r = 0.97$), final viscosity ($p<0.01$, $r = 0.96$) while a significant negative correlation was observed with peak time ($p<0.05$, $r = -0.61$) and pasting temperature ($p<0.05$, $r = -0.69$) of the *eba* (Table 3). Similar correlations were observed in the *Gari* packaged in PVC (Table 4).

Trough viscosity (TV) is the minimum viscosity that occurs after the initiation of product cooling; thus, it measures the ability of the paste to withstand breakdown during cooling. TV values ranged between 210.50 RVU and 383.83 RVU (PP) and 221.21 RVU and 383.83RVU (PVC), which agreed with the value recorded by Sanni et al. (2009) (269.75 RVU). Similarly, the trough viscosity reduced with an increase in the storage periods. The PV of the *Gari* packaged in PP had a significant positive correlation with the trough ($p<0.01$, $r = 0.86$), breakdown viscosity ($p<0.01$, $r = 0.92$), while significant negative correlation was observed with pasting temperature ($p<0.05$, $r = -0.65$) (Table 3). Similar correlations were observed in the *Gari* packaged in PVC (Table 4).

Breakdown viscosity (BDV), which reflected the ability of the sample to withstand shear stress and heating during cooking for this study, was observed to be significantly different ($p<0.05$) during storage and the storage periods had a significant ($p<0.001$) effect on the stored *Gari*. The BDV of this study ranged from 31.55 to 269.79 RVU (PP), and 39.34 to 269.79 RVU for *Gari* packaged in PVC. The highest value BDV was recorded for zero storage, and the value was noticed to decrease with storage periods up to the fourth month of storage; afterwards, the value was higher in the sixth month. The BDV of the *Gari* packaged in PP had a significant positive correlation ($p<0.01$, $r = 0.94$), with a significant negative correlation of the pasting time ($p<0.05$, $r = -0.67$), and pasting temperature ($p<0.05$, $r = -0.69$) of the *eba* (Table 3).

Final viscosity (FV) is the ability of the flour to form starch and viscous paste or gel after cooking and cooling (Maziya-Dixon et al., 2007). The FV ranged from 401.46 to 568.96 RVU (PP) and 400.13 to 568.96 RVU (PVC). There was a significant ($p<0.05$) difference in the FV of *Gari* stored in PVC though, the highest value 503.38 RVU of FV was observed in the second month in the *Gari* stored in PP after zero storage, and this may be ascribed to the packaging materials used (Table 2).

Setback viscosity (SBV) gives an idea about the

Table 3: Pearson correlation of the functional, pasting and sensory properties of *gari* packaged in a polypropylene bag and stored for 20 weeks

	SI	BD	Disp	PV	TV	BDV	FV	SBV	Ptime	Ptemp	Text	Col	Stretch	Mould	Flav	Mouthf	OA
SI	1.00																
BD	−0.11	1.00															
Disp	0.20	0.25	1.00														
PV	0.29	0.43	−0.16	1.00													
TV	0.23	0.54	−0.17	0.96**	1.00												
BDV	0.32	0.31	−0.14	0.97**	0.86**	1.00											
FV	0.14	0.33	−0.37	0.96**	0.92**	0.94**	1.00										
SBV	−0.20	−0.50	−0.49	0.05	−0.17	0.22	0.24	1.00									
Ptime	−0.62*	−0.06	−0.26	−0.61*	−0.48	−0.67*	−0.52	−0.09	1.00								
Ptemp	−0.63*	−0.31	−0.17	−0.69*	−0.65*	−0.69*	−0.56	0.18	0.87**	1.00							
Text	−0.34	0.39	−0.30	−0.03	0.09	−0.12	0.00	−0.21	0.45	0.16	1.00						
Col	0.43	−0.18	0.07	0.03	0.02	0.03	−0.10	−0.28	0.00	−0.05	0.20	1.00					
Stretch	0.24	0.19	−0.18	0.26	0.33	0.19	0.20	−0.30	0.13	−0.14	0.66*	0.53	1.00				
Mould	0.05	0.58*	0.08	0.24	0.34	0.15	0.14	−0.47	0.21	−0.24	0.58*	0.12	0.63*	1.00			
Flav	0.47	−0.29	0.02	−0.03	−0.11	0.03	−0.08	0.07	−0.28	−0.20	0.31	0.54	0.46	−0.04	1.00		
Mouthf	0.56	−0.11	0.21	0.30	0.22	0.35	0.22	0.05	−0.53	−0.63*	−0.09	0.09	0.21	0.37	0.45	1.00	
OA	0.61*	−0.13	0.20	−0.10	−0.16	−0.05	−0.21	−0.14	−0.21	−0.29	0.21	0.50	0.33	0.29	0.78**	0.67*	1.00

*$p<0.05$; **$p<0.01$; SI: Solubility index; BD: Bulk density; Disp: Dispersibility; PV: Peak viscosity; TV: Trough viscosity; BDV: Breakdown viscosity; FV: Final viscosity; SBV: Setback viscosity; Ptime: Peak time; Ptemp: Pasting temperature; Text: Texture; Col: Colour; Stretch: Stretchability; Mould: Mouldability; Flav: Flavour; Mouthf: Mouthfeel; OA: Overall acceptability

Table 4: Pearson correlation of the functional, pasting and sensory properties of *gari* packaged in polyvinyl chloride container and stored for 20 weeks

	SI	BD	Disp	PV	TV	BDV	FV	SBV	Ptime	Ptemp	Text	Col	Stretch	Mold	Flav	Mouthf	OA
SI	1.00																
BD	−0.27	1.00															
Disp	0.52	−0.81**	1.00														
PV	0.13	0.22	−0.53	1.00													
TV	−0.16	0.35	−0.66*	0.91**	1.00												
BDV	0.32	0.10	−0.38	0.96**	0.75**	1.00											
FV	−0.01	0.27	−0.63*	0.97**	0.89**	0.92**	1.00										
SBV	0.30	−0.13	−0.01	0.25	−0.10	0.48	0.38	1.00									
Ptime	−0.54	−0.29	0.24	−0.54	−0.28	−0.66*	−0.47	−0.45	1.00								
Ptemp	−0.65*	−0.27	0.17	−0.54	−0.34	−0.62*	−0.43	−0.24	0.78**	1.00							
Text	−0.032	−0.48	0.58	−0.36	−0.19	−0.44	−0.45	−0.58*	0.35	0.39	1.00						
Col	−0.47	0.14	−0.11	−0.36	−0.19	−0.45	−0.34	−0.35	0.28	0.22	0.30	1.00					
Stretch	−0.19	−0.26	0.27	−0.17	−0.04	−0.24	−0.17	−0.29	0.11	0.12	0.73**	0.37	1.00				
Mould	0.30	−0.21	0.50	−0.26	−0.22	−0.27	−0.29	−0.20	0.05	0.08	0.65*	−0.04	0.54	1.00			
Flav	0.36	−0.22	0.42	−0.11	−0.21	−0.02	−0.140	0.12	−0.20	−0.38	0.35	0.21	0.69*	0.51	1.00		
Mouthf	0.44	−0.18	0.37	−0.06	−0.19	0.03	−0.16	0.04	−0.43	−0.51	0.38	0.35	0.61*	0.39	0.89**	1.00	
OA	−0.12	0.20	−0.06	−0.01	0.08	−0.06	−0.04	−0.25	−0.24	−0.263	0.39	0.43	0.79**	0.32	0.74**	0.75**	1.00

*$p<0.05$; **$p<0.01$; SI: Solubility index; BD: Bulk density; Disp: Dispersibility; PV: Peak viscosity; TV: Trough viscosity; BDV: Breakdown viscosity; FV: Final viscosity; SBV: Setback viscosity; Ptime: Peak time; Ptemp: Pasting temperature; Text: Texture; Col: Colour; Stretch: Stretchability; Mould: Mouldability; Flav: Flavour; Mouthf: Mouthfeel; OA: Overall acceptability

retrogradation tendency of starch in the flour sample after 50 ^{o}C. The SBV values ranged from 125.54 to 190.96 RVU in PP and 132.84 to 194.63 RVU in PVC, and it was observed to decrease with storage periods up until the third month of storage; afterwards, the value was noticed to increase in the fourth month up to the sixth month. However, the highest value was seen in the *Gari* stored in PVC: high SBV values have been reported to affect dough digestibility (Shittu, Lasekan, Sanni & Oladosu, 2001), whilst lower values, which were recorded for third month of storage, are beneficial as they indicate a lower tendency for retrogradation (Sandhu, Singh & Malhi, 2007). The SBV of the *Gari* packaged in PP was negatively correlated ($p>0.05$, $r = -0.49$) with the mouldability of the *eba* but not significant (Table 3) while that of the *Gari* packaged in PVC had a significant ($p<0.05$, $r= -0.58$) negative correlation with the texture of the *eba* (Table 4).

Peak time is reported by Adebowale, Sanni and Awonorin (2005) to be a measure of the cooking time of the flour. The peak times obtained for this study ranged from 5.10 - 6.23 min (PP), and 5.10 - 6.00 min (PVC), which were in line with the studies of Nwancho et al. (2014) (4.67 - 6.47 min) and Awoyale et al. (2016) (5.65 - 5.90 min). Statistically, the storage periods had a significant effect ($p<0.001$) on the *eba* made from the stored *Gari*, while no significant effect ($p>0.05$) of the packaging materials and no combined interactive effect was observed. The results of the peak time of the *Gari* implied that all the *Gari* might be cooked into a paste in <6 mins.

Pasting temperature (PT) is an index of the minimum energy required to initiate rapid water ingression, swelling, and eventual gelatinization of starch granules (Awoyale et al., 2016). Thus, the PT of the *Gari* samples was observed to fall below 100 ^{o}C. The PT of *Gari* ranged from 80.70 - 91.55 ^{o}C (PP) and 80.70 - 87.23 ^{o}C (PVC), which agreed with Awoyale et al. (2016) (78.36 ^{o}C - 80.40 ^{o}C), Olanrewaju and Idowu (2017) (82.05 ^{o}C - 83.66 ^{o}C), but higher than the values reported by Sanni et al. (2008) (63.40 ^{o}C - 64.65 ^{o}C). Similarly, the storage periods had a significant effect ($p<0.001$) on the pasting temperature while the packaging materials and combined interactive had no significant effect on the pasting temperature. The values obtained for *Gari* stored in PP were higher and this was attributed to packaging materials used. However, the high PT value is an indication for ease of formation of paste as reported by Nwancho et al. (2014) and this implies that *Gari* stored in PP tends to spontaneously form a paste of *eba* in contrast to *Gari* stored in PVC.

3.3 Effect of storage conditions and packaging materials on the sensory attributes of cooked *Gari* dough (*eba*)

Sensory evaluation is an expression of an individual likes or dislikes for a product as a result of biological variation in humans and how people perceive sensory attributes. Table 2 shows the sensory attributes of the cooked *Gari* dough (*eba*) produced from *Gari* stored in different packaging materials; PP and PVC. The storage periods and packaging materials had no significant effect ($p>0.05$) on the sensory attributes of the cooked *Gari* dough (*eba*) except for the mouldability, which was significantly ($p<0.05$) affected. The interactions between the storage periods and packaging materials had no significant effect ($p>0.05$) on the cooked *Gari* dough (*eba*) (Table 5). However, the panellists preferred all the sensory attributes of the *eba* from the *Gari* stored in PP compared to the *eba* made from *Gari* stored in PVC. Thus, the overall acceptability was higher in the *eba* made from *Gari* packaged in PP. Therefore, packaging *Gari* in PP may keep most of the properties preferred by the consumers when stored for up to 5 months.

4 Conclusion

The study conducted showed that the packaging materials had no significant effect on the functional properties except for bulk density and all pasting properties of the stored *Gari*, and that the storage periods significantly affected all the functional properties except the swelling power. However, the result of the sensory analysisshowed that during the storage periods only the mouldability and packaging materials had asignificant effect on the sensory acceptability of

Table 5: Effect of storage periods and packaging materials on the sensory acceptability of cooked *gari* paste (*eba*)

Parameters	Storage wks	*Gari* stored in PP Mean	*Gari* stored in PP Range	*Gari* stored in PVC Mean	*Gari* stored in PVC Range	p value of Package	p value of Storage periods	p value of Storage period x Package
Texture	0	7.31±1.97^{a}	2 – 9	7.31±1.97^{a}	2 – 9			
	4	7.54±0.88^{a}	6 – 9	7.69±0.75^{a}	7 – 9	NS	NS	NS
	8	7.62±1.04^{a}	6 – 9	7.23±0.93^{a}	6 – 9	NS	NS	NS
	12	7.54±1.45^{a}	4 – 9	7.46±1.12^{a}	5 – 9	NS	NS	NS
	16	7.69±1.01^{a}	5 – 9	7.77±0.93^{a}	6 – 9	NS	NS	NS
	20	7.54±0.52^{a}	7 – 8	7.54±0.52^{a}	7 – 8	NS	NS	NS
Colour	0	7.69±1.70^{a}	4 – 9	7.69±1.70^{a}	4 – 9			
	4	7.92±0.95^{a}	6 – 9	8.00±0.91^{a}	6 – 9	NS	NS	NS
	8	7.92±0.76^{a}	7 – 9	7.62±0.77^{a}	6 – 9	NS	NS	NS
	12	7.92±0.76^{a}	7 – 9	7.77±0.73^{a}	7 – 9	NS	NS	NS
	16	7.77±0.93^{a}	6 – 9	7.85±0.99^{a}	6 – 9	NS	NS	NS
	20	7.77±0.73^{a}	6 – 9	7.85±0.90^{a}	6 – 9	NS	NS	NS
Stretchability	0	6.31±6.31^{a}	1 – 9	6.31±2.56^{a}	1 – 9			
	4	7.85±0.90^{b}	6 – 9	7.62±1.04^{b}	6 – 9	NS	NS	NS
	8	7.00±1.29^{ab}	4 – 8	7.46±1.27^{b}	5 – 9	NS	NS	NS
	12	7.62±1.61^{b}	3 – 9	7.54±0.88^{b}	6 – 9	NS	NS	NS
	16	7.85±0.90^{b}	6 – 9	7.46±0.97^{b}	6 – 9	NS	NS	NS
	20	7.69±0.85^{b}	6 – 9	7.85±0.69^{b}	7 – 9	NS	NS	NS
Mouldability	0	7.00±2.89^{a}	1 – 9	7.00±2.89^{a}	1 – 9			
	4	7.85±0.80^{a}	6 – 9	7.92±0.95^{a}	6 – 9	NS	*	NS
	8	7.77±1.30^{a}	4 – 9	7.69±1.25^{a}	4 – 9	NS	*	NS
	12	7.85±0.80^{a}	6 – 9	8.00±0.71^{a}	7 – 9	NS	*	NS
	16	8.08±0.86^{a}	7 – 9	7.54±1.12^{a}	5 – 9	NS	*	NS
	20	7.77±1.09^{a}	5 – 9	7.85±0.69^{a}	7 – 9	NS	*	NS
Flavour	0	6.77±1.74^{a}	3 – 9	6.77±1.74^{a}	3 – 9			
	4	7.23±1.01^{a}	6 – 9	7.62±0.77^{ab}	6 – 9	NS	NS	NS
	8	7.38±1.04^{a}	5 – 9	7.46±0.97^{ab}	6 – 9	NS	NS	NS
	12	7.69±0.75^{a}	7 – 9	7.62±0.96^{ab}	6 – 9	NS	NS	NS
	16	7.62±0.96^{a}	6 – 9	7.62±0.87^{ab}	6 – 9	NS	NS	NS
	20	7.62±0.51^{a}	7 – 8	7.77±1.01^{ab}	6 – 9	NS	NS	NS
Mouthfeel	0	6.77±2.00^{a}	1 – 9	6.77±2.00^{a}	1 – 9			
	4	7.00±1.22^{a}	4 – 8	7.46±1.20^{a}	4 – 9	NS	NS	NS
	8	7.23±1.36^{a}	4 – 9	7.38±0.77^{a}	6 – 9	NS	NS	NS
	12	7.85±0.99^{a}	6 – 9	7.62±0.77^{a}	6 – 9	NS	NS	NS
	16	7.31±1.03^{a}	5 – 9	7.38±0.87^{a}	6 – 9	NS	NS	NS
	20	7.31±0.63^{a}	6 – 8	7.46±1.20^{a}	5 – 9	NS	NS	NS
OA	0	7.15±1.82^{a}	3 – 9	7.15±1.82^{a}	3 – 9			
	4	7.77±0.83^{ab}	6 – 9	7.69±0.85^{a}	6 – 9	NS	NS	NS
	8	7.77±0.83^{ab}	7 – 9	7.54±0.78^{a}	6 – 9	NS	NS	NS
	12	7.92±0.64^{ab}	7 – 9	8.00±0.58^{a}	7 – 9	NS	NS	NS
	16	8.08±0.76^{b}	7 – 9	7.85±0.69^{a}	7 – 9	NS	NS	NS
	20	7.77±0.60^{ab}	7 – 9	7.62±0.77^{a}	7 – 9	NS	NS	NS

PP: Polypropylene woven sack, PVC: Polyvinyl chloride container. NS: not significant (p>0.05); *p<0.05, **p<0.01, ***p<0.001, Means in the same row and followed by the same letters are not significantly different from each other (p>0.05) the *eba*. Similarly, *eba* stored in a polypropylene woven sack (PP) gave better sensory properties compared to that stored in polyvinyl chloride containers. Hence, the use of polypropylene bags for packaging and storage of *Gari* is encouraged to retain its sensory attributes.

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Applications of High Pressure Technology in Food Processing

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Abstract

Consumer trends towards shelf-stable, safe, more natural and free from additives foods drove the need to investigate the commercial application of non-thermal food processing technologies. High pressure processing (HPP) is one such emerging technology where foods are generally subjected to high pressure (100-1000 MPa), with or without heat. Similar to heat pasteurization, HPP deactivates pathogenic microorganisms and enzymes, extends shelf life, denatures proteins, and modifies structure and texture of foods. However, unlike thermal processing, HPP can retain the quality of fresh food products, with little or no impact on nutritional value and organoleptic properties. Moreover, HPP is independent of the geometry (shape and size) of food products. The retention of food quality attributes, whilst prolonging shelf life, are enormous benefits to both food manufacturers and consumers. Researches have indicated that the combination of HPP and other treatments, based on the hurdle technology concept, has potential synergistic effects. With further advancement of the technology and its large-scale commercialization, the cost and limitations of this technology will probably reduce in the near future. The current review focuses on the mechanism and system of HPP and its applications in the processing of fruit, vegetables, meat, milk, fish and seafood, and eggs and their derived products.

Keywords: Emerging technology; High hydrostatic pressure; High pressure processing; Non-thermal technology

1 Introduction

Food processing and preservation activities are as old as the human civilization, where foods were generally subjected to roasting, boiling, sun drying, steaming and smoking. Conventionally, most food products are processed thermally (e.g. blanching, drying, baking, evaporation, pasteurization and sterilization) by heating at 60 to 100 °C or more for a few seconds to minutes (James, Martin & David, 1992). During the process, the large amount of energy transferred to the food may trigger detrimental reactions thereby leading to objectionable changes in the food products (Barbosa-Cánovas, Pothakamury, Palou & Swanson, 1998). Thermal treatment of food effectively reduces the number of food spoilage microorganisms but it is necessary to also consider the quality attributes of the material as well as the shelf life. An objective of food industrialists is to develop and implement technologies that can maintain or yield desirable organoleptic characteristics of food or decrease the unwanted changes in commodities due to processing (Hogan, Kelly & Sun, 2005). Therefore, non-thermal methods as an alternative concept or complementary techniques of food processing are being developed and employed. In these methods, the food products are subjected to a lower temperature and less energy is transferred to them compared to thermal processing. The degradation of food quality attrib-

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utes expected from high temperatures is nominal in non-thermal treatment (Hogan et al., 2005). Scientists and researchers are driven to apply the potential of non-thermal technologies as an alternative or complementary process to traditional approaches of food processing and preservation. Food materials can be treated non-thermally using methods such as magnetic fields (MF), high pressure processing (HPP), irradiation, pulse electric field (PEF), pulsed light, ultrasound, ozone, cold plasma, gas and hurdle technology (Pou, 2015).

The potential of high pressure (HP) in food processing was first demonstrated by Hite (1899), with the reporting that milk spoilage by microorganisms can be delayed via the application of high pressure. A high pressure unit, with pressure leak proof sealing, was designed to ensure the system maintained a high pressure. Application of high pressure to food processing is an extension of a technology which is generally used in the manufacture of super-alloys, ceramics, sheet metal forming, low-density polyethylene and simulators. The first commercially high pressure processed food product (jams and jellies) appeared on the market in 1991 in Japan (Yaldagard, Mortazavi & Tabatabaie, 2008). Consumer trends towards shelf-stable, safe, more natural and free from additives foods drove the need to investigate the commercial application of non-thermal food processing technologies. High pressure processing is one such technology where foods are subjected to high pressure (100-1000 MPa), with or without heat (Angsupanich & Ledward, 1998; Rao, Chakraborty, Kaushik, Kaur & Hulle, 2014). Research indicates the great potential to apply high pressure technology in the food industry. Food scientists and technologists reported the various positive applications of HP in the processing/preservation of meat and meat products (Angsupanich, Edde & Ledward, 1999; Cheah & Ledward, 1996; Cheftel & Culioli, 1997; Kaur et al., 2016; Martino, Otero, Sanz & Zaritzky, 1998), fruit and vegetables and their products (Andres, Villanueva & Tenorio, 2016; Arroyo, Sanz & Prestamo, 1997; Cao et al., 2012; Chen et al., 2015; Dajanta, Apichartsrangkoon & Somsang, 2012; De Roeck et al., 2009; Kaushik, Kaur & Rao, 2014; Kaushik, Kaur, Rao & Mishra, 2014; Perera, Gamage, Wakeling, Gamlath & Versteeg, 2010; Rodrigo, Van Loey & Hendrickx, 2007; Sanchez-Moreno et al., 2005), milk and milk products (Addo & Ferragut, 2015; Black, Kelly & Fitzgerald, 2005; Chawla, Patil & Singh, 2011; Naik, Sharma, Rajput & Manju, 2013), eggs (Juliano et al., 2012; Ngarize, Adams & Howell, 2005; Singh & Ramaswamy, 2013), and fish and seafood (Angsupanich et al., 1999; Angsupanich & Ledward, 1998; Kaur, Kaushik, Rao & Chauhan, 2013). The current review of HPP discusses its working mechanism and its applications in the processing of foods.

2 Working principles of high pressure processing

2.1 Isostatic rule

The external pressure exerted on a fluid is distributed evenly and instantaneously throughout the food sample under pressure, whether the sample is in direct contact or indirect (flexible package) contact with the pressure medium irrespective of food geometry and equipment size. This isostatic principle enables the scale-up of laboratory findings to full-scale production of HPP (Olsson, 1995; Rao et al., 2014). When an aqueous medium is compressed, the compression energy can be determined as shown in Equation 1 (Cheftel & Culioli, 1997).

$$E = \frac{2}{5} \times P \times C \times V_0 \qquad (1)$$

Where E is energy (J), P is the pressure (Pa), C is the compressibility of the medium, and V_o is the initial volume (m^3). Thus, the compression energy required to compress 1 litre of water at 400 MPa is 19.2 kJ as compared to 20.9 kJ for heating 1 litre of water from 20 to 25 ^{o}C. Consequently, the low energy levels involved in HPP do not affect the covalent bonds of food constituents (Cheftel & Culioli, 1997).

2.2 Le Chatelier's principle

This principle governs the effect of high pressure on food chemistry and microbiology. When a system at equilibrium is disturbed, the system then responds in a way that tends to minimize the disturbance (Norton & Sun, 2008; Pauling, 1964). In other words, high pressure enhances reactions that result in a decrease in volume (negative activation volume) but opposes reactions that involve an increase in volume (positive activation volume) (Pou, 2015). High pressure reduces the availability of molecular space, favouring the chain interactions and finally inducing negative volume change. An overall volume change enhances the dissociation of ionic interactions and disruption of hydrophobic bonds. High pressure favours the formation of hydrogen bonds, while covalent bonds are not disrupted. Large molecules of microbial cell structures (cell membranes, enzymes, lipids, proteins) are disrupted by HP, while small molecules (flavour components, vitamins) remain unaffected (Rao et al., 2014).

2.3 Heat of compression

Pressure build up (pressurization from P_s to P_1) is accompanied by an increase in temperature (T_s to T_1) through adiabatic heating. During the pressure holding time (P_1 to P_2), the temperature decreases from T_1 to T_2 due to heat loss through the non-insulated pressure chamber as shown in Figure 1 (Balasubramaniam, Ting, Stewart & Robbins, 2004; Rao et al., 2014; Yaldagard et al., 2008). If no heat transfer occurs during the pressure holding time, the product normally cools down to its initial temperature on decompression. The temperature (T_1) at process pressure is independent of the rate of compression provided the heat transfer to the surrounding is negligible. The product temperature increment also depends on material compressibility, specific heat, initial temperature and the pressure requirement. Each product has its own specific heat of compression (fats and oils = 6-8 ^{o}C /100 MPa, water = 3 ^{o}C /100 MPa, 30 % aqueous monopropylene glycol (MPG) = 2 ^{o}C /100 MPa) (Balasubramanian & Balasubramaniam, 2003; Rao et al., 2014) according to its composition.

3 High pressure processing system

The main components of a typical high pressure processing system consist of a high pressure chamber and its closure, a pressure generation system, a temperature control device and a material handling system. The high pressure chamber is the heart of the high pressure processing system, which, in many cases, is a forged monolithic, cylindrical chamber constructed using a low-alloy steel of high tensile strength. The wall thickness of the mono block chamber is determined by the maximum target pressure, chamber diameter and number of cycles (Rao et al., 2014; Yaldagard et al., 2008). The strength of the pressure vessel can be increased by using multilayer, wire-wound or other pre-stressed vessel designs. This type of strengthened pressure vessel is preferred, over a mono block, for safety and reliability in commercial-scale operation at pressures greater than 400 MPa.

The pressurization of food commodities can be achieved by four different approaches, namely, hot isostatic pressing, warm isostatic pressing, cold isostatic pressing and chemical reaction (Mertens, 1995; Rao et al., 2014). In general, high pressure is generated by direct compression, indirect compression and heating of the pressure medium. In direct compression method, the pressure is directly generated by pressurizing a medium with a piston. The large-diameter end of the piston is driven by a low pressure pump. The small-diameter end of the piston pressurizes the pressure medium as shown in Figure 2a. This method allows fast compression, however, the limitations of the high pressure dynamic seal between the piston and internal surface of the pressure vessel confine this approach to small diameter, laboratory or pilot plant systems. Conversely, indirect compression generates pressure indirectly. In this process, a high pressure intensifier is used to force a pressure medium into a closed high pressure chamber from a reservoir through a tubing system until the desired pressure is achieved as shown in Figure 2b. Most industrial operations of the cold and warm isostatic pressing systems employ the indirect compression approach. On the other hand, heating

Table 1: Effect of high pressure processing on fruit and vegetables and their products

Product	Treatment (MPa/°C/min)	Effect	Reference
Guava puree	600/25/15	Extended shelf life up to 40 days stored at 4 °C without any modification in colour, flavour, ascorbic acid concentration, cloudiness and viscosity.	Yen and Lin (1996)
Cauliflower	400/5/30	Induced cell permeability, loss of turgor and structural changes. However, it maintained acceptable flavour and firmness.	Prestamo and Arroyo (1998)
Spinach	400/5/30	Completely destroyed the parenchyma cells and extensively affected the structure.	Prestamo and Arroyo (1998)
Pear	400/20/30	Induced browning, firm texture	Prestamo and Arroyo (2000)
Orange juice	800/25/1	Stabilized fresh orange juice (good cloud stability, lowest level of PME residual activity, less deterioration of ascorbic acid) for a storage period of more than 2 months at 4 °C or 37 °C.	Nienaber and Shellhammer (2001)
Green beans	500/room temperature/1	Extended shelf-life, good firmness, retaining of colour.	Krebbers, Matser, Koets and Van den Berg (2002)
Passion fruit	300/25/5	Not significant change in aroma, flavour, and consistency.	Laboissiere et al. (2007)
Black grape juice	550/44/2	Maximum retention of total antioxidant activity, flavonoids and phenolics.	Chauhan, Raju, Ravi, Roopa and Bawa (2011)
Apricots	300-500/room temperature/5-20	Inactivation of polyphenol oxidase and peroxidase, retention of colour and carotenoids.	Huang et al. (2013)
Apple juice	500/25/3	No significant change in vitamin C content, increase in total polyphenolic content, safe storage for 21 days at 4 °C.	Kim et al. (2012)
Olives	400-600/room temperature/5 and 10	Enhanced shelf-life, no significant change in colour, higher stability and firmness.	Pradas et al. (2012)
Strawberry pulps	400-600/5-25/25	Inactivation of β-glucosidase, polyphenol oxidase and peroxidase enzymes by 41.4, 74.6 and 74.6 % respectively.	Bello, Martinez, Ceberio, Rodrigo and Lopez (2014)
Strawberry	400/room temperature/5	Total anthocyanins content was degraded by 33 % and 57 %, stored at 4 and 25 °C respectively for 45 days.	Gao et al. (2016)
Beet root	650/room temperature/3-30	Up to 25 % inactivation of peroxidase and 10-25 % inactivation of polyphenol oxidase depending on time.	Paciulli, Medina-Meza, Chiavaro and Barbosa-Canovas (2016)
Pear	600/20-100/3-5	Inactivation of peroxidase and polyphenol oxidase by 26 and 68 % respectively, at 20 °C for 5 min. Similarly, 92 and 90 % inactivation at 80–100 °C after 3 min.	Terefe, Tepper, Ullman, Knoerzer and Juliano (2016)
Cloudy apple juice	600/room temperature/3	Up to 50 % inactivation of peroxidase	Yi et al. (2017)

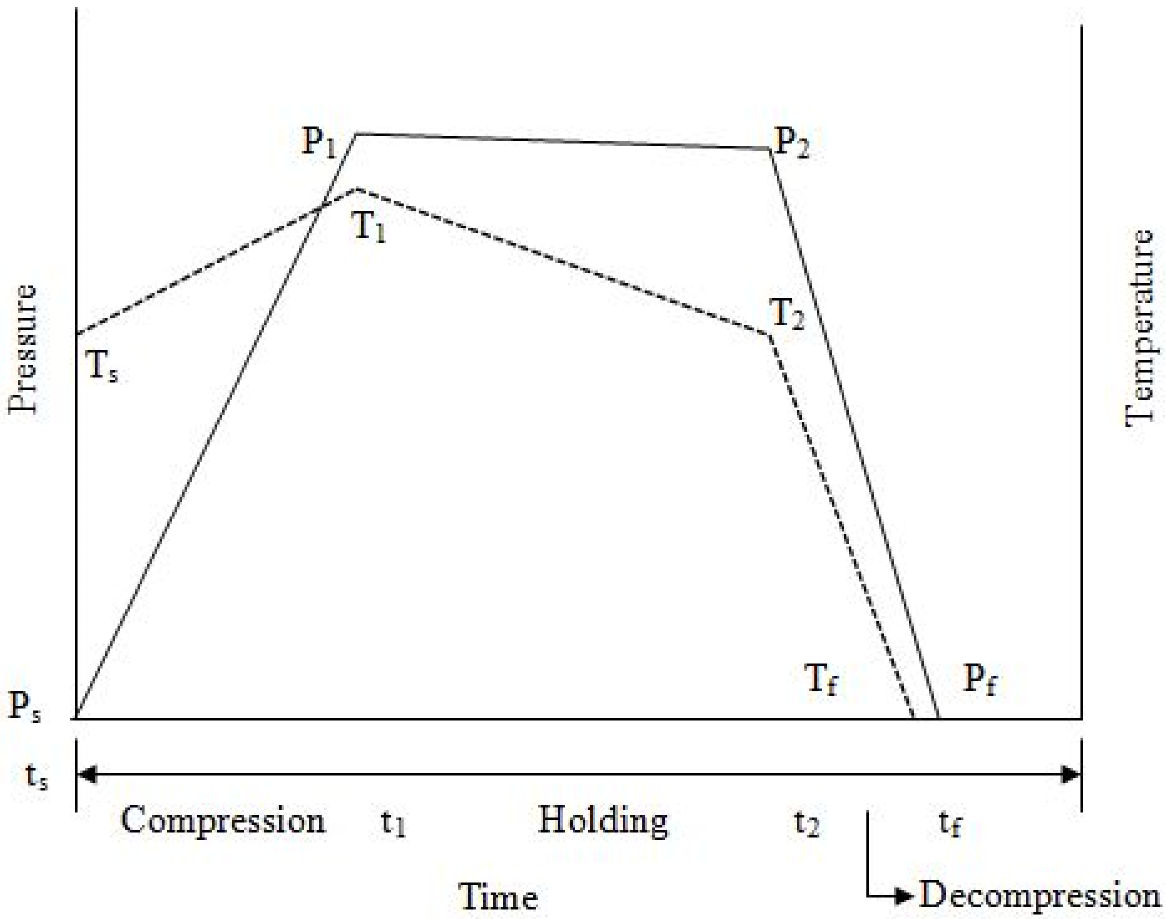

Figure 1: Variation of pressure and temperature in a non-insulated high pressure chamber

of the pressure medium technique utilizes the expansion of the pressure medium with rising temperature to produce high pressure. Therefore, this method is commonly used when high pressure is applied in combination with high temperature (Barbosa-Cánovas et al., 1998; Pou, 2015). Current industrial high pressure modes of operation include batch and semi-continuous systems. The batch mode can process both solid and liquid products. In the batch process, HPP is carried out after food materials are filled and sealed into their final or intermediate package (flexible packaging materials). The advantages of the batch method include freedom from contamination by lubricants and wear particles, and cleaning is not required between food material changes. However, the overall cost of the process is higher as the processing cycle is lengthened due to handling, drying and storage of the packages. In the semi-continuous process, only pumpable products can be used for treatment (Ting & Marshall, 2002). Food products are pumped in and out of the pressure processing chamber by means of special high pressure transfer valves and isolators. The treated foods are packaged using aseptic filling systems (Ting & Marshall, 2002; Ting, Tremoulet, Hopkins & Many, 1999). Pressure transmitting media are used for uniform transfer of pressure to the food products. Some of the most generally used pressure transmitting fluids are water, ethanol, silicone oil, sodium benzoate, glycol and castor oil (Ting et al., 1999). Many of the early pressure processing systems were not fabricated from stainless steel and thus necessitated the use of oils as the pressure transmitting medium. Use of oil as a transmitting medium served the purpose of lubrication and anticorrosion as well as transfer of pressure to the materials. When oils or organic solvents are used as the pressure transmitting medium, the temperature rise is higher than with the use of water, owing to their higher compressibility, lower heat capacity and lower thermal conductivity (Makita, 1992). The main pressure transmitting medium of high pressure in food processing is water because it has the best properties for the process and there is less risk of contamination of the products. Aqueous solutions of mono-propylene glycol (MPG) and isopropyl alcohol (IPA) are also commonly used for high temperature and low temperature high pressure processing, respectively.

4 Biological effects of high pressure processing

4.1 Microorganisms

One of the main objectives of high pressure treatment is the inactivation of food spoilage microorganisms. The inactivation of microorganisms is brought about by the changes in the cell membrane, morphology and biochemical reactions of microorganisms under the influence of HP (Hamada, Nakatomi & Shimada, 1992). The cell membrane of microorganisms is the primary site for pressure induced inactivation of microorganisms, which results in modified permeability and ion exchange. The action of the cell membrane helps the microorganisms to resist some selective chemical inhibitors and maintain homeostasis. However, this tolerance is lost once the cell membrane is damaged and the cells are vulnerable due to high pressure treatment (Manas & Pagan, 2005; McClements, Patterson & Linton, 2001). It is commonly accepted that the reason of cell death is due to the leakage of intracellular constituents through the permeabilized cell membrane. On the other hand, if processing pressure is not high enough to incite total permeabilization of the cell membrane, the permeabilization occurs only in the outer cell membrane and the permeabilized cell is restored upon pressure decompression as in the case of Gram-negative bacteria (Hauben, Wuytack, Soontjens & Michiels, 1996; Yaldagard et al., 2008). The cell membrane fluidity has an effect on the susceptibility of microorganisms to HP. Less fluid cell membrane microorganisms are more sensitive to HP treatments (Macdonald, 1992; ter Steeg, Hellemons & Kok, 1999). High pressure causes the irreversible denaturation of one or more critical proteins in microorganisms, thus leading to the inactivation of microorganisms by altering the proteins responsible for replication, metabolism and integrity. Compression affects the morphology (filament formation, cessation of motility) of the microbes (Kitching, 1957; ZoBell, 1970). High pressure treatment for retardation of reproduction and inactivation of microorganisms is dependent on the types of microorganisms and species, growing stages and level of applied pressure. Cells during the exponential growth phase are more sensitive to pressure than during the stationary phase. In most cases, Gram-negative bacteria are more sensitive to pressure induced inactivation as compared to Gram-positive bacteria. It is well established that spores have higher pressure resistance and for their inactivation high pressures (>1200 MPa) may be required (Knorr, 1995). Generally, pressure treatment at 400-800 MPa for a few minutes at room temperature can satisfactorily achieve microbial reduction. High pressure at around ambient temperature is unfeasible in the inactivation of bacterial endospores. The higher resistivity of spores as compared to vegetative cells is due to the presence of calcium rich dipicolinic acid which defends them from excessive ionization (Sakharam, Prajapati & Jana, 2011; Smelt, 1998). Due to this limitation, high pressure processing is not a suitable method for sterilization; hence, the HP treated products need to be stored under refrigeration. However, HP can stimulate bacterial spore germination, which enables the resultant vegetative form for inactivation by high pressure. Pulsed or oscillatory pressurization, and HP and high temperature combination can enhance the sporicidal effect (Sakharam et al., 2011). Multiple pulse treatment, repeated cycling between 600 MPa and atmospheric pressure, resulted in a 6.0 log reduction of Bacillus stearothermophilus spores whereas single HPP had little effect (Johnston, 1994). The Bacillus stearothermophilus spores were totally destroyed by six 5 min cycle oscillations of pressurization at 600 MPa at 70 ^{o}C (Hayakawa, Kanno, Yoshiyama & Fujio, 1994).

4.2 Enzymes

The basic structure of an enzyme consists of primary, secondary, tertiary and quaternary structures. The tertiary and quaternary structures of an enzyme are affected by high pressure treatment which results from the modification of hydrophobic and electrostatic interactions as well as hydrogen bonding (Marszalek, Wozniak, Kruszewski & Skapska, 2017). The primary structure of an enzyme is unaffected by HP (Heremans, 1993; Mozhaev, Heremans, Frank, Masson & Balny, 1994) whereas the secondary structure of an enzyme may be affected at a pressure greater than 700 MPa. The shift between the native conformations (mostly tertiary and quaternary structures) of the enzyme is dependent on the interaction between the molecules present near the surface and the surrounding solvent molecules. When this balance is disturbed, its conformational structure may change and lead to the loss of its activity (Rao et al., 2014). Effects of HPP on enzymes can be used to enhance some enzyme activities in food to improve food quality or to inactivate the undesirable enzymes using higher pressure. Also, this enhancement of enzyme activity can be used to improve a food process such as cheese production. The mechanism of high pressure inactivation of enzymes can be explained in terms of protein denaturation, complete or incomplete, reversible or irreversible unfolding of enzymatic structure, and influence on the reaction mechanism by altering the difference in reaction volume. It can also be described in terms of modification of the sensitivity of the substrate after being unfolded by the application of pressure, and the bonding of enzyme and substrate may become stronger by the release of an intracellular enzyme (Cheftel, 1992; Ludikhuyze, Van Loey, Denys & Hendrickx, 2001). High pressure inactivation of enzymes can be influenced by type of enzyme, medium, water activity, pH, composition and temperature.

5 Applications of high pressure technology in food processing/preservation

Consumers have a growing interest in safe, healthy, fresh-like, convenient, quality, additive-free, and better texture, flavour and appearance food products. Thermally processed food often results in the deterioration of quality attributes (vitamins loss, change in colour, off-flavour, modification of texture and change in appearance). It is generally accepted that high pressure treatment can inactivate food spoilage microorganisms without having a negative effect on food quality. Increasing pressure level will generally increase inactivation of microorganisms in shorter times but it may also result in more protein denaturation and other unfavourable changes when compared to the untreated food products. However, as high pressure processing generates no shear forces, the physical structure of most processed foods remains minimally changed (Norton & Sun, 2008). The problem of spatial variation is not encountered as the pressure is transmitted evenly and instantaneously throughout the food sample. Moreover, HP affects only non-covalent bonds (ionic, hydrophobic and hydrogen bonds), and has little effect on the quality attributes of food such as nutritional constituents, flavour and colour. Therefore, in contrast with thermal treatment, HPP has a higher potential regarding retention of the inherent food qualities (Hayashi, 1990). Application of high pressure compresses the water content of the food by about 4 and 15 % at 100 and 600 MPa respectively. Freezing point depression of water was observed at HP to -4, -8 and -22 ^{o}C at 50, 100 and 210 MPa, respectively (Kalichevsky, Knorr & Lillford, 1995; Naik et al., 2013). Hence, this method enables sub-zero processing of food without the formation of ice crystal. This technique also assists quick thawing of conventional frozen food products and pressure shift crystallization. In so doing, very small ice

Table 2: Effect of high pressure processing on milk and dairy products

Product	Treatment (MPa/°C/min)	Effect	Reference
Cheddar cheese	345 and 586/5/ 1 and 15	Cheese made from HP treated milk resulted in more yield of cheese and no detrimental effects on flavour. Microbiological quality was comparable to cheese prepared from pasteurized milk.	Drake, Harrison, Asplund, Barbosa-Canovas and Swanson (1997)
Cheese	400/20/20	6.0 log reduction of Penicillium roqueforti	O'Reilly, O'Connor, Kelly, Beresford and Murphy (2000)2
Cheese	500/20/15	Goat milk subjected to HP prior to cheese making gives firmer, less cohesive and less fracturable cheese as compared to pasteurized milk (72 °C for 15 sec).	Buffa, Trujillo, Pavia and Guamis (2001)
Milk	200-500/20/60	Periodic oscillation of HP was observed to be very effective for the inactivation of pathogens such as Escherichia coli, Listeria monocytogenes, and Salmonella enteritidis.	Vachon, Kheadr, Giasson, Paquin and Fliss (2002)
Yogurt milk	200/room temperature/20	Acidification of yogurt milk with glucono-δ-lactone at HP (200 MPa) caused fine coagulum and more homogeneous gel than that of heat treated sample.	Harte, Luedecke, Swanson and Barbosa-Canovas (2003), Naik, Sharma, Rajput and Manju (2013)
Mozzarella and Gouda cheese	400-600/room temperature/5-15	Exposure to HP increased the rate of proteolysis in these cheese varieties. A similar trend was observed in cheese made from the milk of ewe.	Juan, Ferragut, Buffa, Guamis and Trujillo (2007), San Martin-Gonzalez, Welti-Chanes and Barbosa-Canovas (2004)
Cheddar cheese	345-483/room temperature/3 and 7	HP treatment accelerates shredability and shreds from unripe milled curd Cheddar cheese can be manufactured with improved visual acceptability and enhanced tactile handling.	Serrano, Velazquez, Lopetcharat, Ramirez and Torres (2005)
Low fat yogurt	676/85/5 and 30	Combined treatment of HPP and heat using different probiotic starter cultures resulted in higher consistency of yogurt gel, and acceptable textural and rheological properties. It also observed dense aggregated protein structure with smooth surface, and improved viscosity and gel texture as compared to untreated yogurt.	Penna, Gurram and Barbosa-Canovas (2006), Penna, Subbarao-Gurram and Barbosa-Canovas (2007)
Chhana (Indian cottage cheese)	200-400/30-70/0-100	The optimum conditions were determined as 280 MPa, 52 °C, and 45 min for minimum lag, inflexion and coagulation time of 0.0028, 5.19 and 3.87 min, respectively.	Sahu (2010)
Acidified milk gel	200-600/15-65/0-60	The treatment pressure and temperature had the maximum effect on decreasing gel pH. Heat assisted high pressure improved the gel strength and reduced the coagulation time.	Sahu and Mallikarjunan (2016)

Table 3: Some of the effects of HPP on fish and seafood

Product	Treatment (MPa/°C/min)	Effect	Reference
Hake	200 and 400/7/5	Instantly after HP treatment, the samples exhibited odour and appearance similar or somewhat greater than controls samples. During refrigerated storage, the HP treated samples retained all sensory parameters compared to the control samples.	Hurtado, Montero and Borderias (2000)
Pollack, mackerel, tuna, cod, salmon trout, carp, plaice, anglerfish, and octopus.	100-1000/0/5	High pressure treatment higher than 150-200 MPa caused a cooked appearance in all the treated samples except octopus which retained a fresh appearance till 400-800 MPa.	Matser, Knott, Teunissen and Bartels (2000)
Octopus	200-400/7 and 40/15	Samples treated at 200 and 300 MPa showed higher hardness values than both the untreated and treated at 400 MPa samples.	Hurtado, Montero, Borderias and Solas (2001)
Sea bass	100-500/10/5	Chewiness and gumminess decreased from 100-300 MPa but increased after 400 and 500 MPa. A similar trend was observed in hardness but remained constant after 400 and 500 MPa. Springiness, resilience, and cohesiveness remained almost constant.	Cheret, Chapleau, Delbarre-Ladrat, Verrez-Bagnis and De Lamballerie (2005)
Bay scallop	200 and 400/22/10	No modifications were indicated in cohesiveness, adhesiveness, and springiness. Resilience observed to be increased with the pressure level. While hardness decreased regardless of the applied pressure intensity and fracturability found to be decreased at 400 MPa.	Perez-Won, Tabilo-Munizaga and Barbosa-Canovas (2005)
Abalones	500-550/20/3, 5 and 8	Samples treated at 500 and 550 MPa no considerable differences were observed as compared to the control samples. However, chewiness and cohesiveness were higher in the HP treated samples.	Briones-Labarca, Perez-Won, Zamarca, Aguilera-Radic and Tabilo-Munizaga (2012)
Smoked cod	400-600//5 and 10	HP treated samples induced no significant changes during refrigerated storage in the quality of odour, appearance, and intensity of smoky odour.	Montiel, De Alba, Bravo, Gaya and Medina (2012)
Sea bass, sea bass fillets	250 and 400/6/5	HP treated influenced on colour, increased whiteness and appeared typical cooked fish. The brightness was intensified at 400 MPa. Pressure levels did not affect the fresh odour, became firmer, and the overall sensory acceptance was high.	Teixeira et al. (2014)
Threadfin bream	200, 400 and 600/10, 30 and 50	HP processing resulted in a decrease in total sulfhydryl content of actomyosin with the rise in pressure intensity level and treatment time.	Zhou et al. (2014)
Barramundi minced muscle	300-500/4/10	Water holding capacity of gels increased with the increase in pressure intensity. HP treatment enhanced the gel-forming ability.	Truong, Buckow, Nguyen and Furst (2017)

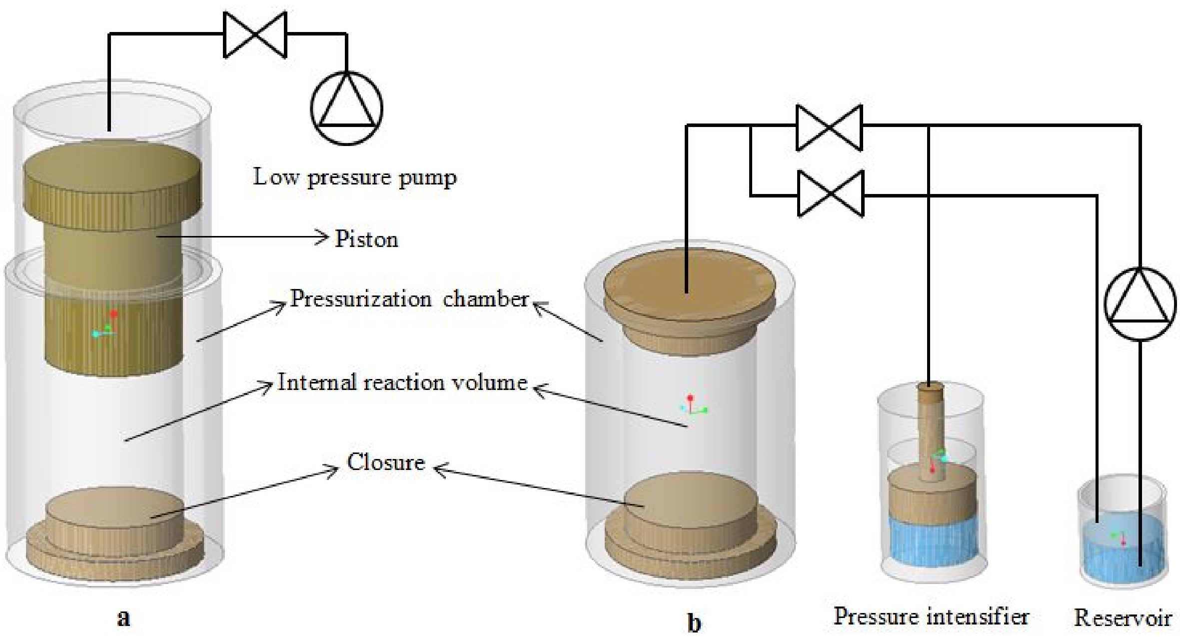

Figure 2: High pressure generation by (a) direct compression, and (b) indirect compression (redrawn from Norton and Sun (2008)

crystals are formed and cooling to sub-zero temperature in the frozen products significantly controls the activities of microorganisms and subsequently improves food quality as well as extending the shelf life of the products (Naik et al., 2013; Sakharam et al., 2011). Bioactive molecules such as simple sugars, vitamins, flavour compounds and amino acids remain unaffected by the application of HPP (Naik et al., 2013).

5.1 Fruit and vegetables and their products

Texture is the most important determinant for the assessment of fruit and vegetables quality. Cell wall polysaccharides are primarily composed of pectin, cellulose and hemicellulose. Pectin is the main component in the middle lamella that strengthens the cell walls and provides elasticity and firmness to the tissues (Kato, Teramoto & Fuchigami, 1997; Rastogi, 2010). Application of pressure can change these compositions, as certain enzymes are deactivated and/or structural modification occurs in the polysaccharide, lipid and protein fraction. The actions of pectinmethylesterase (PME) and polygalacturonase (PG) on pectin caused the texture degradation of fruit and vegetable (Sila, Smout, Vu, Van Loey & Hendrickx, 2005). The action of PME on pectin produced methanol and a pectin molecule with a lower degree of demethylation, which is depolymerized by polygalacturonase, resulting in a drastic softening of tissue (Tangwongchai, Ledward & Ames, 2000; Vu et al., 2004). The rigidity of the cell wall and middle lamella increases due to more crosslinking between pectin chains and divalent cations (like calcium, magnesium) which results from low levels of methoxy pectin (Grant, Morris, Rees, Smith & Thom, 1973; Rastogi, 2010). In some cases, the texture of fruit and vegetable may be enhanced by PME (Villarreal-Alba, Contreras-Esquivel, Aguilar-Gonzalez & Reyes-Vega, 2004). In general, HPP leads to the disruption of membrane and protein denaturation. HP treatment modifies cell permeability and allows the movement of water through the cell. Consequently, HP treated tissue had a drenched or soaked appearance. Yet, some fruit and vegetables retained acceptable firmness, close to that of the original (Rastogi, 2010). Basak and Ramaswamy (1998) reported that texture recovery of some vegetable products was attained between 25 and 40 min under a pressure level of 100 MPa. Tangwongchai et al. (2000) indicated that textural damage of HP processed tomatoes increased with increase in pressure up to 400 MPa at ambient temperature but HP treatment at 500-600 MPa resulted in less damage of texture. The decrease in cell rupture at 500-600 MPa was due to the action of the PME enzyme, and increase in cell rupture and softening below 500 MPa was attributed to the role of the PG enzyme. The texture degradation of high pressure treated mushrooms was found to be lower when compared to thermal blanching (Matser, Knott, Teunissen & Bartels, 2000). High pressure application to green peas (400-900 MPa, 5-10 min, 20 ^{o}C) showed no significant effect on texture (Pandrangi & Balasubramaniam, 2005). The effect of HPP on texture varies with type of fruit and vegetable and applied pressure.

Generally, the colour of HP processed fruit and vegetable products (such as fruit juices, jams, purees) are preserved once thresholds of temperature and/or pH are observed (Ludikhuyze & Hendrickx, 2001). Van Loey et al. (1998) demonstrated that colour degradation of broccoli juice was observed after exposure to pressures at higher temperature (>50 ^{o}C) due to chlorophyll degradation. But below 50 ^{o}C, high pressure up to 800 MPa showed no negative effect on chlorophyll. In the case of onion, with an increase of the applied pressure intensity, the colour turned browner due to the polyphenol oxidase (PPO) enzyme (Norton & Sun, 2008). Thus, the ability to retain colour at HP treatment is not evident in some fruit and vegetables. When treating mango pulp at 100-400 MPa, with the interval of 100 MPa for 15 and 30 min at 20±1.5 ^{o}C, it was found that, after pressure processing, the changes in colour parameters of mango pulp were not significant, indicating minimal effect on pigments. The total colour change decreased with the increase of pressure intensity. The total soluble solids and pH remained unaffected after the high pressure treatment (Ahmed, Ramaswamy & Hiremath, 2005). High pressure (600 MPa) processed cubes of Granny Smith and Pink Lady apples, with pineapple juice 0-50 % (v/v) at 20 ^{o}Bx for 1-5 min at 20 ^{o}C, showed that the HP treatment with 50 % pineapple juice for 5 min resulted in the best quality retention in both varieties of apple. This combination also inactivated the polyphenol oxidase enzyme by 40 % and 30 % in Granny Smith and Pink Lady apples, respectively. Thus, the combined treatment of high pressure and pineapple juice has better possibilities in retention of qualities in both the apple varieties (Perera et al., 2010). Arroyo et al. (1997) have reported that at the pressure level of 100 and 200 MPa at 20 ^{o}C for 10 min and 10 ^{o}C for 20 min, reduction of microbial populations in vegetables (lettuce and tomatoes) were not significant. For the complete reduction of saccharomyces cerevisiae, the required pressure intensity was 300 MPa at 10 ^{o}C for 20 min, and for Gram-negative bacteria and moulds it was 350 MPa. The Gram-positive bacteria were not completely inactivated at 400 MPa. The viable aerobic mesophiles and molds and yeasts were reduced by 1 log unit at 300 MPa and above. High pressure treatment of litchi fruits at 300 MPa for 10 and 15 min, the aerobic mesophiles, yeasts and molds and psychrotrophs count were reduced by 3.29, 3.24 and 3.77 log10 cycles, respectively. The treatment enhanced shelf life up to 32 days with minimal changes in physicochemical attributes and textural parameters during refrigerated storage (Kaushik, Kaur & Rao, 2014). In most cases, during processing and subsequent storage, high pressure treatment can retain higher levels of antioxidant activity and phenolic compounds in fruits and vegetables, and their derived products, than thermal pasteurisation. At present, a significant number of high pressure treated fruit and vegetable products are commercially available in the United States, Europe, Japan and Australia (Zhao, Zhang & Zhang, 2017). It has been scientifically and commercially demonstrated that high pressure processing can produce microbiologically safe and stable fruit and vegetables products, with better quality characteristics. Some of the effects of high pressure treatment on fruit and vegetables and their products are shown in Table 1.

5.2 Meat and meat products

The quality of meat is generally defined by compositional quality and palatability factors. The important quality indicators of meat include colour, flavour, tenderness, juiciness, smell, texture, firmness, fat and protein content, fat quality (oxidative stability of fat), and drip and cooking loss. The processing parameters and methods have effects on the quality attributes of meat that may be beneficial or detrimental. HPP influences the organoleptic properties and nutritive value of meat and meat-derived products since high pressure has a considerable impact on the structure and functionality of many proteins (Jung, de Lamballerie-Anton & Ghoul, 2000; Norton & Sun, 2008). Modification of the ultrastructure of meat is highly dependent on the time post-mortem (pre-rigor or post-rigor) when HP is applied. At early pre-rigor high pressure treatment, the muscles experience great contraction, with a length reduction of 35-50 %, causing severe disruption in the structures of meat (Campus, 2010; Cheftel & Culioli, 1997; Kennick, Elgasim, Holmes & Meyer, 1980). The muscle induced no contraction but modified the sarcomere structure when HP was applied post-rigor, and there is no significant effect on tenderization of post-rigor meat at low temperature (Cheftel & Culioli, 1997; Norton & Sun, 2008). Pressurization up to 500 MPa and above is possible due to advancement in pressurization equipment and may achieve tenderization of meat without any additional heating. The combined effects of high pressure and muscle contraction could result in breakage of myofibrillar proteins, and myosin filaments into Z discs, which would explain the effect of tenderization on meat. The tenderization of meat has also been attributed to the enhanced activity of the enzymes, cathepsins and calpains (Simonin, Duranton & de Lamballerie, 2012). HP treatment (200-500 MPa, 10-30 min at 25 ^{o}C) of beef liver increased the swelling of mitochondria and decreased rough endoplasmic reticula in hepatocytes, and the occurrence of such changes might be related to the modification of texture in the treated beef liver (Ogihara, Suzuki, Michishita, Hatakeyama & Okada, 2017). Banerjee et al. (2017) reported that the high pressure treatment of mutton patties at 200 and 400 MPa for 10 min significantly reduced hardness, gumminess and chewiness as compared to control and irradiated (1-3 kGy) products. However, no significant alterations were observed in springiness and cohesiveness between the HP processed and irradiated mutton patties. Comparatively, HP treated meat batters give rise to more elastic gels during cooking than in cooked-only batters, and reduced cooking losses (Iwasaki, Noshiroya, Saitoh, Okano & Yamamoto, 2006; Sikes, Tobin & Tume, 2009; Truong, Buckow, Nguyen & Furst, 2017). No modifications in meat myofibrils were observed in beef treated at 130 MPa at 10 ^{o}C but when beef was treated at 325 and 520 MPa increasing ultrastructure changes to the meat myofibrils were observed (Jung et al., 2000). High pressure (100 MPa at room temperature) treated pork and chicken induced morphological changes and appeared thicker than untreated samples (Iwasaki et al., 2006; Simonin et al., 2012). Kaur et al. (2016) reported the high pressure processing of bovine meat at 600 MPa induced significant changes in texture, visual appearance and myofibrillar structure, which were similar to cooked meat. However, the sample subjected to 175 MPa exhibited no significant modification in texture and appearance as compared to the raw meat product. In contrast to cooked meat, HP processed meat at 600 MPa was found to have better protein digestibility in terms of free amino N release. However, high pressure processing and cooking are not comparable as the HP effect on meat is an entropy-driven process, whilst cooking is essentially an enthalpy-driven process (Kaur et al., 2016). HP induced modifications in texture have effects on myofibrillar proteins and their gel-developing properties, thus, raising the prospect of the development of treated muscle-based food products (Campus, 2010).

Colour is one of the main quality attributes of meat that consumers use as a purchasing criterion. HP processed meat colour greatly depends on the intensity of the applied pressure, as HP treatment at 130 MPa enhanced redness but above 325 MPa induced strong discolouration (intensifying in brown colouration). This discolouration in meat is due to an increase in the metmyoglobin (Fe^{3+}) content in the sample after the application of pressure (Jung, Ghoul & de Lamballerie-Anton, 2003). Jung et al. (2003) observed that an increase in applied pressure, up to approximately 350 MPa at 10 ^{o}C for 5 min, increased redness (a* values) in raw beef muscle and then decreased redness up to 600 MPa. These authors reported that an increase in redness of samples at pressures below 300 MPa was due to the activation of the enzymatic system accountable for the reduction of metmyoglobin. Marcos, Kerry and Mullen (2010) also observed the decrease in a* values of treated samples at pressures above 350-400 MPa. Dark-firm-dry beef subjected to high pressure at 200 MPa increased the redness value (a*) but treatment at 600 MPa reduced the redness in samples (Utama et al., 2017). High pressure (600 MPa) processed dark-firm-dry beef was found to have higher L* values than those treated at 200 MPa and for the control (Utama et al., 2017). The increase in whitening (L* parameter) has been observed in pork meat processed at 200-400 MPa and 20 ^{o}C (Korzeniowski, Jankowska & Kwiatkowska, 1999), chicken meat treated at 400-500 MPa and 5-10 ^{o}C (Del Olmo, Morales, Avila, Calzada & Nunez, 2010), and beef meat processed at 200-600 MPa and 10 ^{o}C (Carlez, Veciananogues & Cheftel, 1995; Marcos et al., 2010). The meat discolouration in HP processed samples has been related to either (i) a whitening effect due to denaturation of myoglobin and heme group displacement or release, or (ii) oxidation of ferrous myoglobin to ferric myoglobin, or (iii) modification of surface structure and properties due to protein coagulation with a resulting loss of solubility of sarcoplasmic and/or myofibrillar proteins (Campus, 2010; Carlez et al., 1995; Goutefongea, Rampon, Nicolas & Dumont, 1995; Simonin et al., 2012). Comparatively, the colour of raw meat is more affected by pressure than cured meat (Rubio, Martinez, Garcia-Cachan, Rovira & Jaime, 2007). Meat discoloration is not much influenced by the high pressure treatment duration and can be observed after only 1 min of exposure to pressure (Del Olmo et al., 2010). Even though HPP induced visible modifications in colour of raw meat, the colour difference was less perceived after cooking (Mor-Mur & Yuste, 2003; Simonin et al., 2012). High pressure processing has the potential to influence the aroma of treated meat and meat-derived products. The aroma profile of high pressure (400-600 MPa for 15 min at 5 ^{o}C) treated beef and chicken meat had better stability as compared to untreated meat during storage. It was observed that upon opening the bags after 14 days of storage, the untreated samples produced unpleasant off-flavour (Schindler, Krings, Berger & Orlien, 2010). Rivas-Canedo, Juez-Ojeda, Nunez and Fernandez-Garcia (2011) investigated the effect of HP (400-600 MPa for 5-10 min at 12 ^{o}C) on the volatile profile of cooked pork meat and found that the volatile fractions of HP processed meat remained unaltered during the 14 days of refrigerated storage, however, the control samples experienced significant changes. Dry-cured loins subjected to high pressure at 300-400 MPa for 10 min at 20 ^{o}C stabilized the content of free amino acid during storage due to a decrease in the activity of aminopeptidases (Campus, Flores, Martinez & Toldra, 2008). High pressure processing at 200-400 MPa at ambient temperature retained the components (amino acids, nucleotides and peptides) responsible for the flavour of meat during chilled storage for 7 days (Suzuki et al., 1994). Utama et al. (2017) reported that the HP treatment of dark-firm-dry beef at 200-600 MPa induced changes in aroma during 9 days of storage under vacuum at 4 ^{o}C. It was observed that the aroma pattern of untreated meat was always discriminated from HP treated samples during storage. Changes in pH during storage and lipid oxidation products (pentanal and heptanal) might contribute to changes in aroma (Utama et al., 2017). Lipid oxidation is one of the main causes of deterioration of meat and meat-derived products during subsequent storage, particularly cooked poultry and pork meat which contain a significant quantity of unsaturated fatty acids. Lipid oxidation may impair flavour as well as nutritional value (fat soluble vitamins, essential fatty acids). Besides, it may be a health risk as lipid oxidation is linked to the development of cancer and coronary heart diseases (Cheftel & Culioli, 1997). High pressure induces lipid oxidation in meat products, which results in the formation of secondary lipid oxidation products such as thiobarbituric acid-reactive substances (TBARS) and hexanal (Kumar, Yadav, Ahmad & Narsaiah, 2015; Simonin et al., 2012). The oxidation rate of the samples subjected to high pressure can be influenced by the physical treatment conditions, mechanical processing, composition and types of products. In several studies, high pressure induced lipid oxidation was not increased immediately after HP treatment but during subsequent storage of the meat and meat products (Beltran, Pla, Capellas, Yuste & Mor-Mur, 2004; Beltran, Pla, Yuste & Mor-Mur, 2003; Orlien, Hansen & Skibsted, 2000). Conversely, Tuboly, Lebovics, Gaal, Meszaros and Farkas (2003) reported an increase in lipid oxidation immediately after the high pressure treatment. It is generally suggested that high pressure induces lipid oxidation by two mechanisms: (i) increase in release of iron from hemoproteins, and (ii) disruption of the membrane (Kumar et al., 2015). Dry-cured ham processed at 400 MPa, and stored for 39 days in a modified atmosphere with 5 % residual oxygen, observed significantly higher values of TBARS, indicating a reduction in oxidative stability during storage (Andres, Adamsen, Moller, Ruiz & Skibsted, 2006). Utama et al. (2017) observed that processed dark-firm-dry beef treated at 200-600 MPa and held under refrigerated storage resulted in the development of lipid oxidation, with significant differences compared to the control. The authors found that the highest values of TBARS were obtained in samples subjected to 600 MPa, at day 3 of storage. Bajovic, Bolumar and Heinz (2012) established the critical pressure levels (between 300 and 600 MPa) that can induce lipid oxidation in meat. Utama et al. (2017) recommended that the temperature in the pressurization vessel should be maintained below 20 ^{o}C in order to minimize the risk of lipid oxidation in pressure treated meat. High pressure induced lipid oxidation may limit the usefulness of high pressure technology for meat and meat-derived products unless antioxidants are added or oxygen-free packaging is used. Adding carbon dioxide prior to pressure application or removing oxygen may be helpful to avoid HP induced lipid oxidation (Campus, 2010; Cheftel & Culioli, 1997). Lipid oxidation may lead to negative effects on flavour and colour, however, the combination of HPP and metal chelators or antioxidant packaging could reduce the lipid oxidation triggered during high pressure processing (Stratakos & Koidis, 2015). The addition of tomato waste (0.30 %) or final tomato paste (0.10 %) to minced meat resulted in a lag phase of 6 days for the development of secondary oxidation products in the meat subjected to a pressure of 600 MPa (Alves, Bragagnolo, da Silva, Skibsted & Orlien, 2012). Chicken patties packed in antioxidant-active packaging made with a film containing 0.45 mg of rosemary extract /cm^2, treated at 800 MPa for 10 min at 5 ^{o}C and then stored at 5 ^{o}C for 25 days, was able to delay the HP induced lipid oxidation (Bolumar, Andersen & Orlien, 2011). Ethylenediaminetetraacetic acid (EDTA) and egg white powder prevented chicken meat slurries from pressure-induced lipid oxidation (300 and 500 MPa) during chilled storage due to their abilities to chelate metal ions (Beltran, Pla, Yuste & Mor-Mur, 2004; Simonin et al., 2012). One of the main purposes of high pressure processing of meat and meat-based products is to improve microbial safety. The effects of HPP on microorganisms are well recognized and accepted. HPP at low temperature or moderate temperature led to inactivation of enzymes and microbial vegetative cells but was not effective for deactivation of spores (Hugas, Garriga & Monfort, 2002; Stratakos & Koidis, 2015). Different combinations of pressure, temperature, time and cycling treatments can be selected to achieve the complete inactivation of spores (Torres & Velazquez, 2005). On the other hand, comparatively moderate pressure levels of 200-300 MPa are enough to inactivate most food parasites (Lindsay et al., 2008; Porto-Fett et al., 2010; Simonin et al., 2012). Garriga, Grebol, Aymerich, Monfort and Hugas (2004) reported, HP treatment of dry-cured ham, cooked ham and marinated beef loin at 600 MPa for 6 min was found to be an effective method for preventing the growth of Enterobacteriaceae and yeasts. Also *Salmonella* spp as well as *Listeria monocytogenes* were absent during 120 days of storage. Cooked chicken breast subjected to HP at 600 MPa for 2 min at 20 ^{o}C resulted in a decrease of *Listeria monocytogenes* by a 3.3 log reduction (Patterson, Mackle & Linton, 2011). High pressure treatment of beef liver at 400 and 500 MPa for 10-30 min at 25 ^{o}C reduced bacteria by more than 3.0 log, however, the samples subjected to a lower pressure resulted in insufficient microbial reduction for safe consumption (Ogihara et al., 2017). Generally, a higher level of pressure and treatment time led to higher cell reductions (Patterson et al., 2011). However, there are other factors that influence the lethality of high pressure like pH, fat content, water activity (a_w) and the types and growth stages of microorganisms. Cells are most resistant at neutral pH, while the destruction efficiency is decreased at higher or lower values of pH (Huang, Lung, Yang & Wang, 2014). Fat content in meat products can affect the antimicrobial efficiency of high pressure treatment as the fat can have a defensive effect on microorganisms (Huang et al., 2014). Low water activity values of foods can result in a baroprotective effect on microorganisms and thus decrease the inactivation (Hereu, Bover-Cid, Garriga & Aymerich, 2012).

5.3 Milk and dairy products

In general, milk and dairy products are processed at a temperature of 70-145 ^{o}C to inactivate food spoilage microorganisms and to ensure food safety for consumption. However, treatment at high temperature deteriorates the sensory and nutritional qualities of food products as many food nutrients are thermally unstable. To overcome this problem, intensive research has been carried out on the use of high pressure processing as an alternative method to traditional thermal processing of milk and dairy products (Liepa, Zagorska & Galoburda, 2016). Even though milk was the first food product to be processed with high pressure by Hite (1899), thus far, HP treated milk products are not commercially available in the market (Norton & Sun, 2008). HPP influences milk properties, physiochemical properties, constituents and microorganisms present in milk. Rastogi and Knorr (2013) reported that HPP was equally effective for pasteurization in destruction of pathogenic and spoilage microorganisms compared to thermal treatment. Complete destruction of alkaline phosphatase in milk has been observed at 800 MPa for 8 min. Several studies have reported the inactivation of microorganisms either introduced or naturally present in milk. Milk exposed to a microbial 4D HP process at 350 MPa extended shelf life to 12 days at 10 ^{o}C, 18 days at 5 ^{o}C and 25 days at 0 ^{o}C (Mussa & Ramaswamy, 1997). Raw milk subjected to 400 MPa for 30 min at 25 ^{o}C contained less than 7.0 log psychrotrophs/ml after storage at 7 ^{o}C for 45 days, while untreated samples contained more than 7.0 log after storage for only 15 days (Garcia-Risco, Cortes, Carrascosa & Lopez-Fandino, 1998). Raw milk pressurized at 400-600 MPa had a comparable microbiological quality to that of a pasteurized sample at 72 ^{o}C for 15 s, depending on the initial microbiological load of milk samples (Trujillo, 2002). High pressure treated milk at 400 MPa for 15 min or 600 MPa for 3 min at 20 ^{o}C achieved the shelf life of 10 days stored at 10 ^{o}C (Trujillo, 2002). Generally, moulds and yeasts can be destroyed at 200-400 MPa but in the state of a spore or ascospore or in a high sugar concentration food, a pressure of about 600 MPa may be required for inactivation (Bello, Martinez, Ceberio, Rodrigo & Lopez, 2014). Many researchers have reported the inactivation of bacteria in milk at pressures around 400-600 MPa (Amador Espejo, Hernandez-Herrero, Juan & Trujillo, 2014; de Oliveira, Augusto, da Cruz & Cristianini, 2014; Liepa et al., 2016; Patterson, 2005; Udabage et al., 2010).

High pressure has potential to modify the size and distribution of fat globules in milk. On exposure to a high pressure up to 500 MPa, at 25 and 50 ^{o}C, there was an observed tendency to increase the number of small fat globules in the range of 1-2 μm (Gervilla, Ferragut & Guamis, 2001), whilst this tendency was reversed at 4^{o}C. The modification in distribution of fat globules in milk is related to aggregation and disintegration of the fat globule membrane under high pressure. However, there is no damage to the milk fat globule membrane (Dhineshkumar, Ramasamy & Siddharth, 2016). When raw milk was processed at 200 MPa at 4 ^{o}C for 10 or 20 min, the free fatty acids (FFA) content did not alter but slightly increased when treated for 30 min (Kim, Kim, Choi, Min & Kwak, 2008). When ewe's milk was subjected to HP at 100-500 MPa at 4, 25 and 50 ^{o}C, there was no change in the FFA content. In fact, some samples resulted in a lower content of FFA than fresh raw milk when treated at 50 ^{o}C (Gervilla et al., 2001) thus, ameliorating the effects of rancidity in milk during storage. High pressure influenced the colour of milk due to modification in size of fat globules and casein micelles. The optical parameter L* (lightness) of milk exposed to 100-200 MPa slightly differed from the untreated sample whereas those treated at 200-400 MPa saw a progressive reduction (Huppertz, Fox & Kelly, 2004; Huppertz, Kelly & de Kruif, 2006; Needs, Stenning, Gill, Ferragut & Rich, 2000). HP treated skim milk at 200 MPa at 4 ^{o}C was found to decrease L* values (Lee, Choi, Cho & Davaatseren, 2016). Harte, Luedecke, Swanson and Barbosa-Canovas (2003) reported that milk reduced its white colour and changed to yellow when the sample was subjected to thermal treatment followed by high pressure processing (300-676 MPa) but regained its white colour when the sample was exposed to HPP followed by thermal processing. This recovery of the whitish colour may be attributed to the reaggregation of disrupted micelles or the reversible nature of casein micelles. During heat treatment, milk lactose may isomerise in lactulose and consequently degrade to form acids and other sugars. But no modifications in these compounds are detected after the pressure treatment at 100-400 MPa for 10-60 min at 25 ^{o}C, signifying that the Maillard reaction or lactose isomerization did not take place in milk during pressurization (Chawla et al., 2011; Liepa et al., 2016; Lopez-Fandino, 2006). Sierra, Vidal and López (2000) reported that there was no degradation of B group vitamins in HP treated milk. The authors observed no significant loss of vitamin B1 and B6 in milk subjected to high pressure at 400 MPa (2.5 MPa/sec for 30 min at 25 ^{o}C). High pressure processing does not affect the minerals' content of milk but may influence the food matrix leading to an improvement in bioavailability and health benefits (Barba, Terefe, Buckow, Knorr & Orlien, 2015). Milk exposed to HP increases the ionised calcium level, as well as the level of total calcium in the serum phase. The concentration of Ca, Mg and P in serum was found to increase with an increase of pressure to 400 MPa (Barba et al., 2015; Lopez-Fandino, 2006). High pressure induced shifts in the mineral balance leads to an increase in pH of the milk by about 0.1 units. The increase in milk pH and shifts in salts can be reversed rapidly after the treatment of HP, predominantly when the milk is stored at above 10 ^{o}C (Huppertz, Kelly & Fox, 2002; Liepa et al., 2016).

Since high pressure treatment influences the components of milk, it will certainly affect the technological properties of milk during production of various milk products. Milk exposed to 300-400 MPa considerably increased wet curd yield (up to 20 %) and decreased the loss of protein in whey and the volume of whey. The effect is explained by the denaturation of β-lactoglobulin and hence its incorporation in the curd. This results in a high yield of cheese to the extent of 7 %. HP treatment at 400-600 MPa/5-15 min cycle resulted in quick maturation and development of a stronger flavour in cheese (Huppertz et al., 2002). The cheese curd obtained from HP treated milk provides dense network of fine strands, thus showing a great potential for the manufacture of new products owing to the formation of modified textures, tastes and functional properties (Naik et al., 2013). High pressure processing improves the quality and shelf life of yogurt. Exposure of packaged yogurt to high pressure (200-300 MPa at 10-20 ^{o}C) neither changed the texture nor inactivated the viable lactic acid bacteria, however, it prevented acidity development. Above 300 MPa, it resulted in over acidification and reduced viable lactic acid bacteria (Tanaka & Hatanaka, 1992). HP treated (550 MPa) yogurt retained desirable sensory characteristics longer than controls during 4 weeks' storage at 4 ^{o}C or 20 ^{o}C temperature (Jankowska, Reps, Proszek & Krasowska, 2005). Ewe's yogurt made with high pressure processed pasteurized milk (70 ^{o}C for 10 min), using different combinations of pressure and temperature (HP: 200, 350 and 500 MPa and 10, 25 and 55 ^{o}C for 15 min), gave a firmer product on increasing applied pressure. There was a significant increase in gel water retention with the combination of 350 and 500 MPa at 25 and 55 ^{o}C, respectively. Yogurts stored for 20 days at 4 ^{o}C were observed to have good stability (firmness) in all the treatments, however, water retention was only found in yogurt made from high pressure treated milk (Ferragut, Martinez, Trujillo, Güamis et al., 2000; Trujillo, 2002). Yogurt made from HP treated (200-300 MPa at 30 and

40 ^oC) milk was found to delay lipid oxidation and reduce the degree of lipolysis (Dhineshkumar et al., 2016; Serra, Trujillo, Pereda, Guamis & Ferragut, 2008). High pressure also has impacts on dairy products such as ice cream, butter and cream. HPP has the potential for fast ageing of ice cream mixes and the physical ripening of dairy cream for the manufacture of butter. High pressure treatment (300 MPa for 15 min) improved the foaming properties of whey protein concentrate, which improved the body and texture of low fat ice cream when added. The ice cream mix containing HP treated whey protein showed better overrun and foam stability and hardness as compared to ice cream prepared with untreated whey protein. This is due to the influence of high pressure on the functional properties of whey protein (Lim, Swanson & Clark, 2008; Lim, Swanson, Ross & Clark, 2008; Rastogi & Knorr, 2013). Dumay, Lambert, Funtenberger and Cheftel (1996) reported that the pressurization (450 MPa at 10 or 25 ^oC) of pasteurized cream (35 % fat) did not alter the size distribution of fat globules, pH and flow behaviour. It was also observed that there was no further acidification during storage of cream at 4 ^oC for 8 days. On the other hand, HP treatment at 40 ^oC resulted in a modification of fat globules, which is partly reversible with storage. The whipping properties enhanced when cream was subjected to high pressure (600 MPa up to 2 min), possibly due to improved crystallization of milk fat (Eberhard, Strahm & Eyer, 1999; Sakharam et al., 2011). Some of the effects of high pressure treatment on milk and dairy products are shown in Table 2.

5.4 Eggs

Egg products (whole liquid egg or blended liquid egg) are used in a number of food products owing to their high nutritional value and physicochemical properties (such as coagulating, emulsifying and foaming). In general, the quality parameters considered during processing of eggs include texture, microbial inactivation, physicochemical parameters, sensorial quality and shelf life (Wang, Huang, Hsu & Yang, 2016). More than 90 % of food borne *Salmonellosis* is caused by *Salmonella enteritidis* which occurred through egg shell (White et al., 2007). In general, eggs are pasteurized thermally under mild conditions in order to avoid extensive denaturation of proteins. Even the thermal treatment at 60 ^oC for 20-25 min led to partial denaturation of proteins and coagulation, and thus a deterioration in the functional properties of the eggs. Therefore, HPP can be an alternative method to heat treatment as the former has the potential to inactivate microorganisms without adversely influencing the functional characteristics of the egg (Ahmed, Ramaswamy, Alli & Ngadi, 2003; Ponce, Pla, Sendra, Guamis & Mor-Mur, 1999). The impact of HPP on proteins/enzymes and microorganisms has been found to be comparable with that of heating, however, its influence on quality attributes is commonly considered to be minimal. Texture is generally accepted as one of the main sensory characteristics in determining overall quality. Egg white behaves differently to egg yolk and whole liquid egg due to its high protein content (Singh & Ramaswamy, 2013). HP treatment triggered the coagulation of egg white, and an increasing level of applied pressure and treatment time induced the gelation of egg white similar to that of an egg patty. High pressure processing of egg (600-900 MPa for 0-15 min) resulted in full set egg gels with enhanced physicochemical characteristics and without any cooked flavours. Egg white became opaque at 600 MPa and was able to form egg gels at a treatment of 600 MPa for 15 min. Egg yolk and whole liquid egg were able to form gels at HP treatment of 700 MPa for 15 and 10 min, respectively (Singh & Ramaswamy, 2013). Liquid whole egg exposed to a pressure of 150 MPa for 60 min at 25 ^oC was unable to coagulate but at a pressure higher than 250 MPa and a treatment temperature up to 45 ^oC instantaneous coagulation was observed (Lee, Heinz & Knorr, 1999). Egg yolk formed a soft and adhesive gel at HP processing of 400 MPa for 30 min at 25 ^oC. The hardness of the gel increased and adhesiveness reduced with an increase in pressure (Farr, 1990). Similar observations were also found by Singh and Ramaswamy (2013), where HP induced gels were highly elastic and soft. Cohesiveness and hardness of all the egg components were observed to increase with increasing applied pressure, and the increase in egg yolk was greater than in other egg components. The springiness of whole liquid egg was higher compared to egg white and egg yolk, with increased springiness at higher pressure intensity and treatment time. Aguilar, Cordobes, Jerez and Guerrero (2007) reported that rising pressure caused a dramatic alteration in the linear viscoelastic behaviour, undergoing a sol-gel transition. High pressure treatment was also investigated as a function of pH and solids contents. The impact of HPP on aggregation and network development can be modulated by pH to a great level by changing the balance between hydrophobic and electrostatic interactions. HPP (400-600 MPa) caused sufficient modifications in the viscosity of egg components so as to form a gel with enhanced quality attributes as compared to thermally induced gels. Gels formed by HP have been observed to be more elastic and softer without any cooked flavour and taste, and no formation of lysinoalanine and destruction of vitamins have been detected (Hayashi, Kawamura, Nakasa & Okinaka, 1989; Singh & Ramaswamy, 2013).

HPP is being investigated to improve shelf life and reduce the detrimental impact of the pasteurization process. Anton, Chapleau, Beaumal, Delepine and de Lamballerie-Anton (2001) found that high pressure can be used to extend the shelf life of egg yolk based emulsions while reducing the number of microbial counts without modifying their physicochemical properties. Multi-pass HP processing (100 MPa) of whole liquid egg, inoculated with 4.0 and 7.0 log CFU/ml *Salmonella enterica* serovar Enteritidis, was found to exhibit first order inactivation kinetics (Patrignani et al., 2013). *Salmonella enteritidis* inoculated in whole liquid egg was efficiently inactivated after a high pressure treatment at 300-450 MPa for 5-15 min at various temperatures of 15, 20 or 50 ^{o}C (Lai et al., 2010; Ponce et al., 1999). Application of high pressure (300 MPa for 3 min) followed by heating (52 ^{o}C for 3.5 min or 55 ^{o}C for 2 min) of whole liquid egg in the presence of 2 % triethyl citrate resulted in a similar microbial quality level to that of whole liquid egg processed at 71 ^{o}C for 1.5 min but the functional properties remained as those of untreated whole liquid eggs (Monfort et al., 2012). The whole liquid egg subjected to high pressure at 300 MPa for 3.3 min had a reduction of the total microbial count of 1.6 to 3.8 log CFU/g. Also, the authors found that the addition of antimicrobial agents (nisin, monolaurin, lysozyme and EDTA) and subsequent pressure treatment resulted in the reduction of microbial count on average by 3.0 log. The synergistic effect was observed if the antimicrobial agent nisin (or monolaurin) was combined with high pressure (Schenkova et al., 2009). Egg patties exposed to high pressure and temperature (700 MPa and 105 ^{o}C) resulted in the inactivation of *Bacillus stearothermophilus* spores (Rajan, Pandrangi, Balasubramaniam & Yousef, 2006). Koutchma, Guo, Patazca and Parisi (2005) also reported that egg patties treated at 700 MPa for 4 min at 105 ^{o}C led to inactivation of *Bacillus stearothermophilus* spores by 6.0 log whilst *Clostridium sporogenes* PA 3679 was reduced by 6.0 log at 700 MPa for 5 min at 110 ^{o}C. The effects of HPP on the physicochemical properties and structure of ovotransferrin concentrate induced an increase in the surface hydrophobicity of protein without any modifications in the total sulfhydryl groups, hence, aggregation was inhibited. This would be of great help in the development of a microbiologically safe high quality product (Acero-Lopez, Ullah, Offengenden, Jung & Wu, 2012; Wang et al., 2016).

High pressure processing triggered a number of changes in the colour parameters (L*, a*, and b* values) of all the egg components (whole liquid egg, egg yolk and egg white). Singh and Ramaswamy (2013) investigated the effect of HPP (600-900 MPa for 0-15 min) on the colour parameters of egg components. For egg white, the L* (brightness), a* (redness) and b* (yellowness) values were found to increase with a rise in pressure intensity level and treatment time. On the other hand, egg yolk changed its colour from pale yellow to orange yellow whilst L* values remained unchanged and a* values were observed to decrease. However, b* values increased considerably indicating an increase in the yellow colour of the egg yolk. In the case of whole liquid egg, all the colour parameters (L*, a*, and b* values) increased significantly with an increase in applied pressure and time. The increased lightness and reddish yellow colour implied the whole liquid egg was more attractive. When whole liquid egg was exposed to 300 MPa for 3.3 min, the L* value remained unchanged whereas a* and b* values were found to decrease (Schenkova et al., 2009). However, the total colour difference (ΔE*) of HP treated samples immediately after HP treatment and after 7 days of refrigerated storage was found to be lower when compared to pasteurized samples (65 ^{o}C for 3 min), indicating that the HP treated whole liquid egg colour retention was relatively similar to that of the fresh untreated samples. Egg yolk subjected to high pressure (400 MPa for 30 min) retained its original colour (Farr, 1990).

5.5 Fish and seafood

Seafood is exceedingly perishable and post-mortem modifications follow rapidly compared with other muscle foods. It is because of its high water activity, pH close to neutral, unsaturated fatty acids content as well as free amino acids and active autolytic enzymes, thus prone to oxidative and microbial degradation (de Oliveira, Cabral Neto, Rodrigues dos Santos, Rocha Ferreira & Rosenthal, 2017). Chemical tests and total viable counts such as analysis of total volatile basic nitrogen (TVB-N) and trimethylamine (TMA-N) have been used to assess the spoilage of seafood in the seafood industry. The value of TMA-N below 15 mg/100 g and TVB-N less than 300 mg/100g indicates good quality seafood (Ali, Sharif, Adhikari & Faruque, 2009; Kaur et al., 2013). A pressure intensity level of 100-600 MPa for a few seconds to 10-15 min are the most commonly used treatment conditions. With high pressure treatment (200-400 MPa) it was possible to effectively avoid microbial growth, trimethylamine development and autolytic activity in sliced raw squids (Gou, Xu, Choi, Lee & Ahn, 2010). HP processed (250 MPa for 5 five min at 3 ^{o}C and 250 MPa for 10 min at 25 ^{o}C) cold smoked salmon was acceptable for up to eight weeks of storage, hence the shelf life was improved by 2 weeks compared to untreated products (Erkan et al., 2011). Hurdle technology of high pressure processing (250 MPa for 2 two min and 200 MPa for 2 two min) followed by mild heat treatment (45 ^{o}C for 15 min and 50 ^{o}C for 5 five min) of oysters reduced *Vibrio vulnificus* and *Vibrio parahaemolyticus* to non-detectable levels, and retained the sensory characteristics for an extended shelf life (Ye, Huang & Chen, 2012). Black Tiger Shrimp extended its shelf life to 15 days compared with 5 days in untreated samples during chilled storage when treated at 435 MPa for 5 min at 25 ^{o}C (Kaur et al., 2013). Fish are highly susceptible to oxidation due to their high content of polyunsaturated fatty acids, pro-oxidants such as enzymes and transition metals and heme-containing protein such as hemoglobin. The influence of high pressure processing on lipid oxidation depends on various factors such as high pressure intensity, treatment time, ante-and post-mortem, fat profile, age, pre-processes, chemical composition, fibre type and age among others (Truong, Buckow, Stathopoulos & Nguyen, 2015).

A stronger catalytic oxidation power at a pressure level of 300 MPa was reported, even though higher levels of thiobarbituric acid (TBA) were detected from 150-300 MPa in salmon, carp, bonito fish, cod, sea bass, and mahi-mahi (Angsupanich & Ledward, 1998; Lakshmanan, Patterson & Piggott, 2005; Medina-Meza, Barnaba & Barbosa-Canovas, 2014; Sequeira-Munoz, Chevalier, LeBail, Ramaswamy & Simpson, 2006; Teixeira et al., 2014; Wada & Ogawa, 1996; Yagiz, Kristinsson, Balaban & Marshall, 2007). Conversely, smoked fish showed more stability to lipid oxidation after pressure treatment, most likely due to antioxidants derived from the smoke (Jo et al., 2014; Montiel, De Alba, Bravo, Gaya & Medina, 2012). Kaur et al. (2013) reported that the HP treatment of black tiger shrimp at 100, 270 and 435 MPa for 5 min at 25 ^{o}C did not significantly change the free fatty acids (FFA) content, indicating that HP did not affect the hydrolysis mechanism of fatty acids. Similar results were observed in mackerel, turbot and salmon (Chevalier, Le Bail & Ghoul, 2001; Figueiredo, Bragagnolo, Skibsted & Orlien, 2015; Ortea, Rodriguez, Tabilo-Munizaga, Perez-Won & Aubourg, 2010). On the other hand, horse mackerel exposed to HP at 150, 300 and 450 MPa for 0, 2.5 and 5.0 min exhibited an increase in the concentration of free fatty acids, with a significant correlation to HP intensity level and treatment time (Torres, Vazquez, Saraiva, Gallardo & Aubourg, 2013). Sequeira-Munoz et al.

(2006) also revealed a similar phenomenon in carp pressurized at 100-200 MPa for 15 and 30 min. The increase in concentration of FFA may be explained by the unfolding of myofibrillar proteins and interference of interactions between free fatty acids and these proteins triggered by high pressure. The myofibrillar proteins and FFA interact with each other through van der Waals, electrostatic, hydrogen bonding and hydrophobic forces, which result in a decrease of protein extractability (de Oliveira et al., 2017).

Several studies have shown that high pressure processing impacts on the structure and texture of fish and seafood. HP treatment of sea bass fillets (100-500 MPa for 5 min at 10 ^{o}C) showed that above 300 MPa fish were harder after chilled storage as compared to untreated samples, indicating the ability of HP to enhance the textural quality of fish fillets (Cheret, Chapleau, Delbarre-Ladrat, Verrez-Bagnis & De Lamballerie, 2005). Liang, Guo, Zhou, Xiao and Liu (2017) indicated the processing of bighead carp surimi gels at 100-500 MPa for 30 min at 25 ^{o}C exhibited less hardness and chewiness but greater gels' strength than traditional two-step heat treated gels when treated above 300 MPa. The highest gel springiness and strength were found at 500 MPa. They also observed that the adductor muscle of bay scallop decreased hardness as compared to the control samples after HP treatment. Barramundi minced muscle subjected to HP (300-500 MPa for 10 min at 4 ^{o}C) and subsequent cooking (90 ^{o}C for 30 min) increased hardness, springiness, gel-forming ability and water holding capacity, with an increase of pressure level and salt concentration (Truong et al., 2017). HP treatment at 2 % salt concentration developed barramundi gel with greater gel strength, mechanical properties and smoother texture than heat induced gels (90 ^{o}C for 30 min) (Truong et al., 2017). The HP treatment of black tiger shrimp (100, 270 and 435 MPa for 5 min at 25 ^{o}C) resulted in a hardening effect with an increase of the applied pressure (Kaur et al., 2013). Similar trends were observed in tuna, salmon, cod, mahi-mahi and trout (de Oliveira et al., 2017; Ramirez-Suarez & Morrissey, 2006; Yagiz et al., 2007; Yagiz et al., 2009). Modifications in texture can be directly linked to the influence of high pressure on proteins such as protein denaturation and aggregation, denaturation of myofibrillar proteins, α-actinin release and alterations in the actin-myosin interaction (Guyon, Meynier & de lamballerie, 2016; Yagiz et al., 2007).

It is important to understand the effects of HP on fish colour, as colour is one of the main attributes for considering freshness, perception of product quality and influence on the purchase decision of consumers. Several studies have reported that the L* values increase in HP treated fish, which appeared more clear, typical of cooked meat characteristics and grey when exposed to 150-300 MPa (Cheret et al., 2005; de Oliveira et al., 2017; Jo et al., 2014; Truong et al., 2015; Yagiz et al., 2007). HP processed black tiger shrimp gave a significant increase in L* parameters with increased pressure intensity. It was also observed that a* value decreased while the b* value increased after HP treatment (Kaur et al., 2013). Barramundi minced muscle subjected to high pressure resulted in a substantial increase in whiteness, with rise in pressure level (Truong et al., 2017). Similar observations were found in HP treated bighead carp surimi gels (100-500 MPa for 30 min at 25 ^{o}C), where the L* values were observed to increase and a* parameters decrease, with the increase of applied pressure. The b* values were observed to decrease at lower pressures; however, it varies at 300 MPa or greater (Liang et al., 2017). The total colour change (ΔE) tends to increase with high pressure processing as revealed in most of the studies (salmon, carp, cod, oyster, bluefish, mahi-mahi, sea bass, tuna, turbot, shrimp) (de Oliveira et al., 2017). Even though there are some variances between results, most investigations indicate a decrease in redness (a* value) and increase in yellowness (b* value), which differs with high pressure treatment conditions and species of fish and seafood. The colour changes induced by HP might be due to denaturation of globin and/or release or displacement of heme (Cheftel & Culioli, 1997; Kaur et al., 2013). Sequeira-Munoz et al. (2006) proposed that the coagulation of sarcoplasmic and myofibrillar proteins induced by high pressure were responsible for the modifications of colour parameters in the samples. An additional possible cause suggested for the change in colour is lipid oxidation due to degradation of the major carotenoid pigment which results in the release of Fe and Cu ions from the muscles (Cruz-Romero, Kerry & Kelly, 2008; de Oliveira et al., 2017; Kaur et al., 2013). Some of the effects of high pressure processing on fish and seafood are shown in Table 3.

6 Some drawbacks of high pressure technology in food processing

Whilst there are a number of countries worldwide such as USA, Japan, France, Romania, Greece, Belgium, Spain, Portugal and Netherlands manufacturing HPP food products (Bajovic et al., 2012; Rastogi, Raghavarao, Balasubramaniam, Niranjan & Knorr, 2007), HPP use is still limited to a comparatively small number of countries. One of the main limitations of HP applications at the present time is the cost of this technology (including the cost of investment and maintenance of equipment, and limited production throughput due to a discontinuous process) (Stratakos & Koidis, 2015; Zhao et al., 2017). The HPP phenomenon is based on compression, so the food must contain water. High pressure treatment may not inactivate spores and some enzymes are very resistant to pressure. Foods that are structurally fragile require special attention (Naik et al., 2013). Another downside is the restriction on selection of packaging materials, as HPP requires flexible/soft packaging materials and is thus only limited to plastic. The limited large-scale commercialization of HP technology is also caused by the difficulty in fabrication of pressure vessels that will endure the very high pressures required. In addition, repeatedly compressing and decompressing may damage the air-tight HP body and pressure adding container. Also, lack of regulatory approval has hindered a larger implementation of this technology on an industrial scale (Wang et al., 2016). Consequently, considering some of the above difficulties, food manufacturers may prefer conventional methods of food processing/preservation over high pressure technique.

7 Conclusions

High pressure is an emerging non-thermal technology which can accomplish food safety standards comparable to those of heat pasteurization. High pressure processing can destroy pathogenic microorganisms and enzymes, extend the shelf life and change structures, with little or no influence on the nutritional and sensory quality attributes of food products. Several studies have shown the great potential of high pressure processing/preservation of meats, fruits, vegetables, seafood, eggs, milk and their derived products. The combination of HPP and other processing methods (thermal, antimicrobial, antioxidant, metal chelators, vacuum packaging, chilled storage, non-thermal methods, among others) can be suitably selected for the effective treatment of foods. High pressure processing may not replace traditional methods of food processing but it may complement such methods. The future application of high pressure technology in food processing/preservation is promising with the advancements in development of high pressure machinery.

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