

**EVALUATION OF OPTIMAL MALTING CONDITIONS OF
THREE RICE VARIETIES FOR IMPROVED BREWING
TRIALS**

BY

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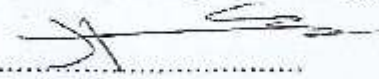
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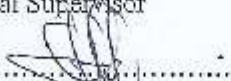
CERTIFICATION

I certify that this work, "Evaluation of Optimal Malting Conditions of Three Rice Varieties for Improved Brewing Trials" was carried out by Akosim, Chibago Queeneth (Reg. No. 20134869468) in partial fulfillment for the award of the degree of Master of Science (M.Sc) in Food Science and Technology (Food Processing Technology Option), in the Department of Food Science and Technology of the Federal University of Technology, Owerri.



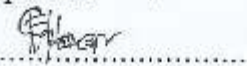
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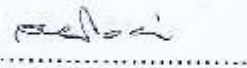
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DEDICATION

I dedicate this research work to God Almighty, my parents Mr. Romanus and Patricia Akosim, My Sponsors Surv. M.C and Dr Mrs T.I Egbe and my daughters Glenda and Gianna Ndubuisi.

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ABSTRACT

Gluten-free beer could be produced with rice, although the latter would primarily serve as an adjunct in combination with barley malt in today's brewing. But, the recent growing realisation of the potentials and applications of rice malt for brewing an all-rice malt beer through varying malting conditions cannot be overlooked. In this current study, therefore, the characteristic changes in malt, wort and beer from different Nigerian rice varieties (FARO 44, FARO 57, NERICA 7) as influenced by varying malting conditions (steeping duration [18, 24 and 30 h], germination periods [2, 3 and 4 days] and kilning temperatures [50 and 55 °C]), were investigated. Rice (grain) samples were examined by thousand corn weight Germinative Energy, Germinative Capacity, and Degree of Steeping. To ensure that rice wort/beer of unique beer style and enhanced attributes comparable to barley wort/beer is produced, only malting conditions that delivered rice malts with peak Diastatic Power, Cold Water Extract and Hot Water Extract were selected. Peak Diastatic Power, Cold Water Extract and Hot Water Extract were obtained at FARO 44 [18h steeping, 3days germination, 55°C kilning (S₁₈G₃K₅₅°)], FARO 57 [30h steeping, 2days germination, 50°C kilning (S₃₀G₂K₅₀°)] and NERICA 7 [24h steeping, 3days germination, 55°C kilning (S₂₄G₃K₅₅°)]. Selected malts were further tested for Moisture Content, Total Nitrogen, Malt Yield, and Malting Loss, which subsequently progressed to wort and beer production. Wort's pH, Total Soluble Nitrogen, Brix, Kolbach Index, Free Amino Nitrogen, Dextrose Equivalent, Original Extract, and Sugar Profile were determined, as well as beer's pH, Colour, Apparent Extract, Alcohol by volume (%ABV), Turbidity, and Sensory attributes. Rice grains' Thousand Corn Weight, Germinative Energy, Germinative Capacity, and Degree of Steeping significantly varied ($p < 0.05$) across varieties. Despite wort's pH, Total Soluble Nitrogen, Dextrose Equivalent, Original Extract as well as beer pH, Colour, Apparent Extract, and Turbidity resembled ($p > 0.05$) across varieties, the wort's Brix, Kolbach Index, Free Amino Nitrogen, Sugar profile as well as beer's %ABV, differed significantly ($p < 0.05$). Sensory attributes of appearance, colour, mouthfeel, and overall acceptability in beer differed noticeably ($p < 0.05$), except for both aroma and taste ($p > 0.05$). Overall, the rice beer, though very slightly hazy, represented a pale yellow light lager, which suggested a peculiar beer style. In addition to benefits of increased Diastatic Power and enhanced hydrolysis, varying malting conditions could be key in reducing the cost of exogenous (commercial) enzymes or barley malt imports, together with decreasing barley's dependency for brewing in the tropics.

Keywords: malting conditions; rice malt; rice beer; rice wort; mashing process.

CHAPTER ONE

INTRODUCTION

1.1 Background to the Study

Global production of rice (*Oryza sativa*) to meet up with consumer demand is projected to potentially double by 2050 (USDA-ERS, 2019; OECD/FAO, 2019). In Africa, Nigeria leads in rice production (Daoui, 2018) with 2018 rice paddy production record of 6.81 million tonnes (World Data Atlas, Accessed 08 June 2020). The recent curb to imported rice was aimed to allow for increased local production (Russon, 2019). Nigeria imported barley malt until the 1988 ban (Koleoso and Olatunji, 1992), which intensified focus of indigenous breweries on locally produced/commercially thriving cereals like maize, rice, and sorghum. Besides rice, barley is another global cereal that breweries utilize (Contreras-Jimenez *et al.*, 2018, Daneri-Castro, *et al.*, 2016). Non-temperate countries unable to commercially produce barley, supplement by imports (either malted or unmalted) for their breweries.

Prior to the 1988, Nigeria barley malt importation ban however, various pilot plant and commercial tests had established locally cultivated sorghum malt or grit as brewing candidates compared with existing barley malt brands (Koleoso and Olatunji, 1992). Resembling those of sorghum, malted maize brewing properties could potentially replace the barley malt (Okafor and Aniche, 1980). By assessing malting and brewing potentials, Okafor and Iwouno (1991) reported Nigerian rice varieties as promising substitute for barley in beer

production. Odibo, *et al.*,(2002) reported fermentable extracts from locally cultivated sorghum to keep for a longer time, until required in the brewing process. Whilst Ogbeide (2011) showed sorghum as an adjunct to malted barley in wort production/brewing process, Iwouno and Ojukwu (2012) showed malting quality potential of a Nigerian locally cultivated yellow maize variety. Recently, Ofoedu, *et al.*, (2019) reported sugar profile of local rice product/derivate (syrup) resembled that of barley wort.

Malting, within the brewing process, is employed particularly to prepare the brewing raw material. The next step that subsequently follows is the mashing and wort fermentation. It is important to reiterate that the malting process involves steeping, germination, and kilning. This process is very crucial in beer production because it helps to develop/prepare the inactive hydrolytic enzymes present in the raw grain (Dewar,*et al.*,1997). Steeping enhances grain softening, increases water availability, and stimulates germination (Sripriya,*et al.*,1997). Mashing, however, facilitates enzymatic degradation of polysaccharides present (in malt), which eventually converts to alcohol in fermentation step of beer manufacturing (Gupta,*et al.*, 2010).Moreover, it has been understood that the low protein and fat content of rice have the potential to assure a slightly higher starch content of 80 to 90 % (Wani *et al.*, 2012; Omar *et al.*, 2016; Othman and Omar, 2017) compared to barley starch content of approximately 70 % (Asare *et al.*, 2011; Zhu, 2017). This aspect of rice might suggest a higher extract yield (Narziss and Back, 2012), though with different starch structure, and

composition in amylose and amylopectin, as well as a lower amylolytic activity than barley (Cela *et al.*, 2020). Therefore, there is a need to optimize the malting and mashing conditions of rice. Moreover, rice has been reported to yield incomplete saccharified mash (wort) (Teeravivattanakit *et al.*, 2017; Roberto *et al.*, 2020) which could be due to its insufficient inherent starch-degrading enzymes (low diastatic power) and high gelatinization temperature (Cela *et al.*, 2020), owed to the nature of rice starch.

Applying such processing methods as malting on rice can enhance the degradation (depolymerization) of its high molecular weight constituents (starch and proteins) to achieve lower molecular weight constituents (sugars and amino acids), which could eventually influence its composition and functionality during processing (Shumin *et al.*, 2014). This opinion appears to concur with the findings of previous workers (Mayer *et al.*, 2017; Usansa *et al.*, 2011) wherein, despite the lower diastatic power of rice compared to barley, rice can serve as a raw material candidate for brewing given its higher limit-dextrinase content compared to barley that elevates the complete saccharification of rice wort, provided the malting conditions are optimized. In addition, the high fiber content of rice assures an enhanced lautering process, attributable to its filtering capacity (Kongkaew, *et al.*, 2012). The sufficient (structural) protein degradation either prior to or simultaneously with starch saccharification, according to Narziss and Back (2012), can expose the grain cell wall structure, thus enhancing the easy breakdown of its endosperm (Shumin *et al.*, 2014;

Kohorn, 2000). As the protein degradation appears more challenging in the malted rice compared to barley, the endogenous enzyme in the rice malt would facilitate the breakdown of rice constituents *in vivo* and improve the extract yield in the wort production. During the malting process, additionally, the rice would attain a higher alpha-amylase production (Ayernor and Hammon, 2000) together with a suitable beta-amylase activity (Cela *et al.*, 2020). Besides starch and protein degradation of rice, the actions of malting conditions enhance enzyme production, which aids *in vivo* hydrolysis of endosperm in the rice kernel during germination and *in vitro* hydrolysis during mashing (Hassani,*et al.*, 2013; Garzon,*et al.*, 2016). Varying the malting conditions, to ascertain the situation that would bring about rice malts with potentially higher diastatic power, and fermentable extracts appears a promising remedy to the brewing challenges associated with rice.

Nigeria's local rice varieties, having evolved over the years with improved qualities like (longer) grain length, improved colour/cooking quality, etc, continues to compete with foreign ones, thrives increasingly, and spreads its distribution/reputation to the West African sub-region. Besides the increased interest to find local/indigenous raw material(s) to supplement barley in brewing, to help reduce (barley) imports, increase (local/indigenous) rice production (Index Mundi, 2020), Nigeria's quest to attain self-sufficiency (in rice production) is not far-fetched, as have been described elsewhere (Ofoedu *et al.*, 2020). Placing the greater emphasis on such underutilized indigenous rice

varieties, particularly those perceived as undesirable due to widely accepted factors such as poor cooking quality (soft and sticky grains), poor physical attributes (poor colour, short-grain length, etc.), and poor consumer acceptability, therefore, makes (rice) product quality diversification should be very fitting. The growing realization of researchers, (rice) processors, and local breweries about the potentials as well as the diverse applications of these (underutilized indigenous) rice varieties, have recently facilitated (rice) product diversification, to actualize promising products like syrups (Ofoedu *et al.*, 2020), gluten-free beers (Cela *et al.*, 2020), flours, and malts (Osuji *et al.*, 2019). Additionally, even though rice grits have served as adjuncts in brewing, the use of rice malt as a specialty ingredient or base malt in the brewing industry, for instance, an all-rice malt beer (gluten-free rice beer) (Marconiet *al.*, 2017) should be very promising. Additionally, the malted rice specifically harnessed from locally produced indigenous rice varieties in Nigeria appears not fully explored, particularly as a principal raw material or substrate for brewing.

1.2 Statement of the Problem

Barley which is the principal grain in brewing is a temperate crop. Countries outside the temperate regions do not grow barley in commercial amounts, and hence, depend on importation from the temperate countries, resulting to depletion of foreign exchange; therefore, there is need for a cheaper source of locally available raw material. The use of rice in brewing has posed a great difficulty due to incomplete saccharification caused by poor modification of

endogenous enzymes during malting. Utilization of exogenous enzyme to achieve complete saccharification of wort is an integral part of mashing operation but expensive as well. Therefore, there is need to reduce the cost of enzyme used during mashing by optimizing malting conditions that will enhance development of adequate endogenous enzyme in the grist.

1.3 Objectives of the Study

The Main objective of this research work is to evaluate the optimal malting conditions of three rice varieties for improved brewing trials.

The Specific objectives are

1. To explore malted rice from indigenous rice varieties as a principal raw material for brewing.
2. To modify malting and mashing conditions of three rice varieties and evaluate the malting characteristic.
3. To evaluate the sugar profile, quality characteristics of the wort and beer.
4. To evaluate the sensory qualities of the trial beer produced from the malted rice varieties.

1.4 Justification of the Study

The success of this work has inaugurated a new frontier and will encourage more research for barley malt replacement and reveal more possibility of using tropical cereals (rice) in beer brewing.

The information obtained from malting trials will help to reveal that complete saccharification can be achieved when malting conditions are modified to optimize enzymes for brewing.

Utilizing tropical grains such as rice of different varieties will also reduce the percentage of our country's foreign earnings spent on importation of barley.

This research work will guide the Nigerian government to enlighten and encourage farmers to increase production of these tropical cereals thereby creating more employment for these farmers and at the same time encourage more food technologists and local enterprises to venture into tropical beer brewing.

1.5 Scope of the study

The scope of this work covered modifying the malting conditions (steeping duration, germination days and kilning temperature) of three rice varieties in order to determine their malting and grain quality characteristics as well as the malting condition with higher cold-water extract, hot water extract and diastatic power performance. The work also covered utilization of the selected malting conditions of different rice varieties for brewing trials. Some physicochemical analysis (pH, total soluble nitrogen, brix, kolbach index, dextrose equivalent original extract and free amino nitrogen) and sugar analysis were carried out on the wort. Sensory analysis and quality evaluation (pH, colour, apparent extract, alcohol by volume and turbidity) were also carried out on the beer from

different rice varieties and the data obtained from these determinations were subjected to statistical analysis.

CHAPTER TWO

LITERATURE REVIEW

2.1 Cereal crops

Africa with its vast land area covering 3 billion ha has 1.3 billion ha of agricultural land out of which only 252 million ha (19.36 %) is arable (2011, FAO). Africa is the center of origin and a major producer of several cereals like sorghum, pearl millet, finger millet and African rice. Another major cereal, maize, has overtaken these traditional cereals while wheat is widely cultivated in North Africa, Sudan and Ethiopia. Agriculture is the ‘engine for growth’ in Africa. With subsistence agriculture practiced by majority small holder farmers, yield gaps are high and poor soils, amongst other constraints add to the difficulties for sustainable farming and incomes. Cereals like Sorghum, Millets, Wheat, Maize and Rice are major staple foods of the most population. These cereals are grown over an area of 98.6 m ha producing 162 m tons.

Maize is a major staple food crop grown in diverse agro-ecological zones and farming systems and consumed by people with varying food preferences and socio-economic backgrounds in sub-Saharan Africa (SSA). The central role of maize as a staple food in SSA is comparable to that of rice or wheat in Asia, with consumption rates being the highest in eastern and southern Africa (ESA). Of the 22 countries in the world where maize forms the highest percentage of

calorie intake in the national diet, 16 are in Africa. Maize accounts for almost half of the calories and protein consumed in ESA, and one-fifth of the calories and protein consumed in West Africa. An estimated 208 million people in SSA depend on maize as a source of food security and economic wellbeing. Maize occupies more than 33 million ha of SSA's estimated 200 million ha of cultivated land. Considering the low average maize grain yields that are still pervasive in farmers' fields meeting the projected increase demand for maize grain in Africa presents a challenge.

Sorghum is the second most important cereal after maize with 22% of total cereal area, followed by millets (pearl and finger) with 19% of the total cereal land coverage. The continuing demand for these two crops is reflected in the trend for increasing area under sorghum and millets in Africa over the last fifty years. Unfortunately, however, crop productivity has not kept pace with increasing demand, due mainly to a lag in crop improvement efforts in sorghum and millets, relative to other cereals, and the extreme environmental conditions and resource constrained, low-input farming systems where these crops are grown. Furthermore, in such dryland environments, the issues of climate variability, change and land degradation are acute with a lack of progress the result of neglect remoteness and weak national institutions. Despite these factors there is a strong case for stepping up the efforts towards development of technologies (germplasm improvement, agronomic management), markets and

institutions to advance the case for sorghum and millets in the dryland tropics of Africa.

Rice (*Oryza sativa* L.) is a cereal which forms an important part of the diet of a great population worldwide for at least 5,000 years. It is native to tropical and subtropical southern Asia, and southeastern Africa. Rice is widely grown throughout the world. Total rice production for 2004 is placed approximately 600 million metric tons per year. Thailand is the world class rice exporter, which produces more than 30 million metric tons of rice per year and highly diversity of rice variety approximately 85 cultivars have been promoted for plantation. Rice consists of cover grain in which the kernel (caryopsis), which is encased in a protective hull.

The hull has two-piece structures, which tightly encompasses the kernel for protect and the barrier to change in moisture content rapidly (Hettiarachchy *et al.*, 2000).

After the hull is removed, the caryopsis is known as brown rice which approximately 93% endosperm is situated. The major constituent of the endosperm is starch and the outer most layer of endosperm called aleurone layer. The area between hull and endosperm composes of three layers: pericarp, tegmen (seed coat), and nucellus account for approximately 3% of brown rice weight.

Rice is currently the second most widely used as adjunct material in United State less than corn grits. The quality of rice for brewing can be judged by

several factors including cleanliness, particle size, gelatinization temperature, mash viscosity, mash aroma, and protein content. Short grain rice is preferred because of the low viscosity problem. However, rice has relatively high gelatinization temperature (70 - 85 °C) due to the presence of very small starch granule (2-10 µm) (Lindeboom *et al.*, 2004). Rancidity of rice grain due to the unfavorable storage condition such as high temperature and humidity, high lipid content of rice could be cause of the reduction (1973). Furthermore, rice provides a low foam quality, stability, and free amino nitrogen (FAN), because of the level of proteins and enzymes.

Rice has become a highly strategic and priority commodity for food security in Africa. Consumption is growing faster than that of any other major staple on the continent because of high population growth, rapid urbanization, and changes in eating habits (Seck *et al.*, 2013). It is the single most important source of dietary energy in West Africa and the third most important for Africa as a whole. Although local rice production increased rapidly after the 2007-2008 food crises, a key problem facing the rice sector in Africa in general is that local production has never caught up with demand. The continent therefore continues to rely on importation to meet its increasing demand for rice. Wheat is grown on around 10 million ha in Africa. It is a major staple crop for several countries and an imported commodity in all of Africa. In all African countries, wheat consumption steadily increased during the past 20 years because of growing population, changing food preferences and socioeconomic change associated

with urbanization. African countries are the world's biggest wheat importer with more than 45 m t in 2013 at around 15 billion US\$. Wheat imports account for 60% of African's wheat consumption and 80% of Sub-Saharan (SSA) countries. North African countries have the highest per capita wheat consumption and wheat provides up to 50% of daily calories and protein. In rapidly urbanizing sub-Saharan Africa, wheat consumption is expected to grow 38% by 2023 with imports already at 23 m tons of wheat in 2013 at a cost of \$7.5 billion. Considering the growing importance wheat has for food security in Africa, African Union Heads of State endorsed their Agriculture Ministers' endorsement in January 2013, to add wheat to the list of strategic crops for Africa.

Cereal yields in Africa are lower than half the world average. The average fertilizer consumption is 16.24 kg/ha (2010, FAO) which is 1/6th compared to the world consumption of 98.20 kg/ha. Increasing productivity of the small holder farmers, bridging the yield gaps by providing appropriate inputs along with improved technologies such as stress resistant and high yielding varieties will be a step towards agricultural transformation in Africa.

Africa faces a wide range of challenges in the production of the five major cereals considered in this Work Stream - rice, maize, millet, sorghum, and wheat. Key among these challenges are:

A.The impact of climate change: Global agriculture is facing the probable impact of global warming. Recent studies suggest that the production of major

commodities has declined since 1980 due to global warming (Lobell *et al.*, 2011). It is estimated that, given current warming trends in sub-Saharan Africa, the production of major cereals could decline by as large as 20% by mid-century (Schlenker and Lobell 2010). The poor who depend on agriculture for their livelihoods and are less able to adapt will be disproportionately affected (World Bank 2007). A recent study estimates the annual costs of adapting to climate change in the agricultural sector to be over USD 7 billion (Nelson, 2009).

B. Land degradation and persistent biotic and abiotic stresses: In addition to inherently high climate variability, the looming threat of higher temperatures and more vicious droughts (arising from climate change) is a major concern. Further, high incidences of diseases, insect-pests, and parasitic plants, and sub-optimal soil nitrogen have also presented a continuous challenge to cereal productivity in SSA3.

C. Rapid population growth across Africa and associated difficulty in meeting the projected demand for food: The high population growth in Africa is giving rise to rapidly increasing demand for food. The UN's Human Development Indices suggest that the dryland areas in West and Central Africa (WCA) and East and Southern Africa (ESA) remain among the poorest and most food-insecure places in the world.

D. Poor mechanization: The low level of mechanization in African agriculture has continued to serve as a huge impediment towards advancing cereal

production, especially of wheat and rice which, in turn, results in the high cost of producing these crops.

E. Inadequate or weak policy environment: Most government policies are inappropriate and inconsistent, and do not provide an enabling environment for the development of the cereal sector in Africa. This includes low funding of the national agricultural research and extension institutions, leading to ineffective technology development and diffusion mechanisms. Lack of investment in infrastructure such as roads, storage and market facilities handicap the potential role of the private sector.

F. Dwindling financial resources for Research and Development: There has been a steady decline in the level of financial support by the major donors to agricultural research, over the last 5 years. Many CGIAR centers have had to cut back their financial allocations to these cereals.

2.2 Description of Rice Caryopsis

The rice grain (rough rice or paddy) consists of an outer protective covering, the hull, and the rice caryopsis or fruit (brown, cargo, dehulled or dehusked rice), (Juliano and Bechtel, 1985). Brown rice consists of the outer layers of pericarp, seed-coat and nucellus; the germ or embryo; and the endosperm. The endosperm consists of the aleurone layer and the endosperm proper, consisting of the sub aleurone layer and the starchy or inner endosperm. The aleurone layer encloses the embryo. Pigment is confined to the pericarp (Juliano and Bechtel,

1985). The hull (husk) constitutes about 20 percent of the rough rice weight, but values range from 16 to 28 percent. The distribution of brown rice weight is pericarp 1 to 2 percent, aleurone plus nucellus and seedcoat 4 to 6 percent, germ 1 percent, scutellum 2 percent and endosperm 90 to 91 percent (Juliano, 1972).

The aleurone layer varies from one to five cell layers; it is thicker at the dorsal than at the ventral side and thicker in short-grain than in long-grain rice's protein bodies, containing globoids or phytate bodies, and in lipid bodies. The aleurone and embryo cells are protein bodies, containing globoids or phytate bodies, and in lipid bodies (Tanaka *et al.*, 1973; Tanaka *et al.*, 1977).

The endosperm cells are thin-walled and packed with amyloplasts containing compound starch granules. The two outermost cell layers (the sub aleurone layer) are rich in protein and lipid and have smaller amyloplasts and compound starch granules than the inner endosperm. The starch granules are polyhedral and mainly 3 to 9 μm in size, with unimodal distribution. Protein occurs mainly in the form of spherical protein bodies 0.5 to 4 μm in size throughout the endosperm (Bechtel and Pomeranz, 1978), but crystalline protein bodies and small spherical protein bodies are localized in the sub aleurone layer. The large spherical protein body corresponds to PB-I of Tanaka *et al.*, (1980) and the crystalline protein body is identical to PB-II. Both PB-I and PB-II are distributed throughout the rice endosperm.

Non-waxy rice (containing amylose in addition to amylopectin) has a translucent endosperm, whereas waxy (0 to 2 percent amylose) rice has an

opaque endosperm because of the presence of pores between and within the starch granules. Thus, waxy grain has about 95 to 98 percent the grainweight of non-waxy grain.

2.3 Chemical Composition of Cereals

In barley malt brewing, the process of solubilization (enzymatic hydrolysis and physical solubilization) of the chemical components of the grain: starch, non-starch polysaccharides (NSPs), proteins, lipids, minerals, vitamins and phytochemicals is facilitated by enzymatic modification of the grain structure during malting. Notably, the endosperm cell walls are degraded and there is limited hydrolysis of the endosperm protein matrix and starch granules (Morral and Briggs.,1978). The tropical cereal grains are similar in general proximate chemical composition to barley with only a few clear differences. However, there are important detailed differences in the composition, structure and properties of some of the chemical components between them and barley, and between themselves, which can restrict solubilization even if these grains are malted. The starch gelatinization temperature range of the tropical cereal starches is some 10 to 20 degrees higher than that of barley starch, 62-78°C and 51-60°C, respectively (Lineback, 1984). The temperature optima of, for example, sorghum malt α -amylase is around 70°C and sorghum malt β -amylase is around 50°C (Kumar *et al.*, 2005; El Nour *et al.*, 2010), with complete inactivation of β -amylase taking place at 68°C within 15 minutes (Taylor and Robbins, 1993). Because of this, simultaneous starch gelatinization and

hydrolysis which takes place in barley malt mashing (Briggs *et al.*, 2004) is not effective. Thus, in practice the tropical cereals, whether in the form of raw grain or malt, must be cooked first to gelatinize their starch, cooled then saccharified using barley malt or commercial enzymes (Briggs *et al.*, 2004; Taylor *et al.*, 2006). Thus, in commercial lager brewing using sorghum malt, the malt is primarily an adjunct rather than the source of hydrolytic enzymes (Taylor *et al.*, 2006). This is despite much research on rising temperature and decoction type mashing regimes to obviate the problem of sorghum starch's high gelatinization temperature (Igyoret *et al.*, 2001). There is some evidence that the gelatinization temperature of sorghum and rice starches may be generally slightly higher than that of maize starch (Hoseney, 1994).

In the case of sorghum, this is possibly because the amylose degree of polymerization (DP) and average number of side chains appears to be higher than that of maize starch (Taylor and Emmambux, 2010). However, the proven, considerably higher gelatinization temperature of tropical starches seems to be related to the longer side chains of their amylopectin compared to those of the temperate cereals.

Rice is unusual in that its starch is stored in compound starch granules consisting of at least 16 small granules of 3-5 microns' diameter (Fitzgerald, 2004). However, the compound granule structure does not appear to affect end-use quality (Fitzgerald, 2004). Stickiness of long grain rice is apparently correlated with the proportion of amylopectin A and short B chains

(Cameron and Wang, 2005). There also exist waxy cereals, which are essentially 100% amylopectin (0% amylose) maize (Watson, 2003), rice (Wilkinson and Champagne, 2004) and sorghum types. The high proportion of amylopectin exerts considerable effects on starch physical properties. Waxy barley and maize starches have been found to exhibit much greater swelling than their normal counterparts, which had 27.5% and 29.4% amylose, respectively. Similarly, waxy sorghum starch exhibited a very much higher and considerably earlier pasting peak viscosity than normal sorghum starch, despite its gelatinization temperature being two degrees higher (Sang *et al.*, 2008). Presumably because of the better swelling properties of amylopectin starch, there has been substantial interest in using waxy sorghum and maize in lager beer brewing (Ortega *et al.*, 2004).

2.3.1 Protein content

Quantitatively, the major proteins of barley, maize and sorghum are the prolamin storage proteins, which are endosperm-specific (Hoseney, 1994). Uniquely, in rice glutelin-type storage proteins with an 11S globulin type amino acid sequence (Yano *et al.*, 2001) are the major proteins. Whole grain rice also has much lower protein content (approx. 7%) (Shih, 2004) than the other cereals barley (8-15%), maize (approx. 10%) and sorghum (approx. 11%) (Schake *et al.*, 2004). The maize zein and sorghum kafirin prolamins are very similar in composition, amino acid sequence and conformation, consisting predominantly of small, 19-25 kDa alpha-prolamins (Belton *et al.*, 2006) and hence are less

diverse than the barley hordein proteins, which more closely resemble the wheat gliadins and glutenins (Bekes and Wrigley, 2004). Kafirin and zein are also notably more hydrophobic, or more strictly speaking less hydrophilic, than the storage proteins of the triticeae cereals such as wheat and barley (Duodu *et al.*, 2003) Importantly with respect to the provision of free amino nitrogen (FAN), the wet cooked protein digestibility of sorghum is substantially lower (approx. 30% lower) than that of the other cereals. Having said this, there is also some evidence that the endosperm storage proteins of cooked rice are very resistant to hydrolysis. Concerning the low protein digestibility of cooked sorghum, this is because of extensive polymerization of the kafirins (and possibly other endosperm proteins) through disulphide bonding involving the cysteine-rich γ -kafirin sub-class (Ng'andwee *et al.*, 2008; Da Silva *et al.*, 2011). The cross-linking of the kafirin containing endosperm matrix protein may limit starch gelatinization during cooking (Ezeogwe *et al.*, 2008) and in turn hydrolysis of the starch to fermentable sugars. The disulphide-bonded cross linking involving γ -kafirin seems to exacerbate the problem of the high content of proline in cereal prolamin proteins, for example γ -kafirin 23 mole% proline (Shewry, 2002). The high content of proline makes the prolamin oligopeptides, released by endopeptidase activity, resistant to degradation to free amino acids by conventional carboxypeptidases.

Rice is a model plant for genomic and proteomic studies, due to its small genome compared with other cereals. In recent years, there has been a significant

progress in the identification and cataloging of rice proteins. More than thirteen thousand proteins, expressed in different tissues and organelles, have been detected, out of which 5,755 have been classified and had their functions determined (Komatsu, 2006). Information on the rice proteome is available in databases such as the Rice Proteome Database (National Institute of Agricultural Sciences, 2002). However, there is little information on grain storage proteins in these databases (Komatsu, 2006). Storage proteins accumulate in large quantities during seed development and are mainly stored in special organelles called protein bodies (Halford and Shewry, 2002). As shown by (Kim *et al.*, 2009), the total storage protein content in *O. sativa* endosperm varies between 4.3 and 18.2%. These storage proteins are divided into four fractions according to their differences in solubility: albumins (soluble in water), globulins (soluble in salts), prolamin (soluble in alcohol), and glutelin (soluble in acidic or basic solutions). Glutelin fraction is the predominant protein in rice endosperm, and is classified according to the molecular weights of α -glutelin (37 kDa) and β -glutelin (20 kDa) (Katsude-Tanaka *et al.*, 2004). Rice protein is considered to be of good quality because it contains eight out of ten essential amino acids. Compared with other cereals, such as maize and wheat, rice has a high level of lysine, which provides high digestibility and nutritional quality (Huebner *et al.*, 1990). Determining the contents of amino acids in storage proteins is an excellent source of information for planning the development of rice genotypes with higher grain nutritional quality. In Brazil,

rice accounts for 14% of the energy and 10% of the protein consumed daily by the population (Naves and Bassinello, 2006).

2.3.2 Non-starch polysaccharides (NSP)

A fundamental difference in structure and chemical composition of rice compared to sorghum and maize is that paddy (rough) rice has a fibrous husk (hull) like barley, which is not threshed off the true grain. The rice hull comprises some 31-36% cellulose, 18% pentosans, 10-18% lignin, 3-12% hemicelluloses and 13-21% ash (mainly silicon)(Champagne *et al.*, 2004). Rice hulls could presumably function as a filter bed in lautering.

The cell walls of the starchy endosperm, like the starchy endosperm matrix protein, may limit starch granule expansion during cooking and subsequent starch hydrolysis. Also, importantly the endosperm cell walls can retard or limit wort separation due their hydration. As is well known, this can be a major problem in barley brewing where the water-extractable (1-3,1-4) β -glucans, are by far the major cell wall component. Rice endosperm cell walls seem to be different to either barley or sorghum and maize, as although arabinoxylans and β -glucans account for the major proportion (approx. 47-49%) of endosperm cell wall NSP (Fincher and Stone, 2004).

2.3.3 Lipids

In brewing, lipid content is important in the foam development and stability of beer (Briggs *et al.*, 2004). Whole grain maize has a considerably higher fat content (approx. 4.4%)(Morrison, 1993), compared to barley (2.3-

3.7%)(Watson, 2003), barley malt(approx. 1.8%)(Schakel *et al.*, 2004), rice (1.6-2.8%) (Schakel *et al.*, 2004) and sorghum (approx. 3.2%) (Serna-saldivar and Rooney, 1995). The lipid content of the endosperm tissue is much lower.

2.3.4 Phenolics and tannins

All cereal grains contain phenolic acids, which are concentrated in endosperm and bran cell walls and most cereals contain some flavonoid- type polyphenols, which are concentrated in the pericarp (Dykes and Rooney,2007). However, sorghum is unique among the major cereals in that certain sorghum varieties contain significant levels of condensed tannin type polyphenols ($\geq 1\%$) and that most types contain significant levels of flavonoids (Dykes and Rooney, 2006).

2.4 Rice cultivation and consumption

Nigeria's rice sector has witnessed some remarkable developments particularly in the last ten years. Both rice production and consumption in Nigeria have vastly increased during this period. The demand for rice in Nigeria is, however, growing faster than for any other major staples, with consumption broadening across all socio-economic classes, including the poor. Substitution of rice for coarse grains and traditional roots and tubers has fueled growth in demand at an annual rate of 5.6 per cent between 1961 and 1992 (Osiname, 2002). FAO (2003) projected growth in rice consumption for Nigeria beyond year 2000 remained as high as 4.5 per cent per annum. In response to the growing demand for this staple, government at various periods actively interfered in the rice

economy coming up with policies and programs one of which is the enormous (rice) imports to supplement the local production which no doubts constitute an enormous drain on the countries had earn foreign earnings.

Others had included oscillating import tariffs and import restrictions. Notwithstanding the various policy measures, domestic rice production has not increased sufficiently to meet the increased demand.

Rice is cultivated in virtually all the agro-ecological zones in Nigeria. Estimate of locally produced rice for year 2002 was 2.9million tones (FAOSTAT, 2005). Also, only about 6.7 per cent of the 25 million hectares of land cultivated to various food crops was cultivated to rice between 2000 and 2002(Osiname, 2002). Paddy rice production in Nigeria has not followed any clear-cut pattern but seems to oscillate with policies of various governments. The trend in production shows that a boom was first experienced in 1965-1970 periods when average output stood at 321 thousand tons. During this period, average area cultivated to rice stood at 234 thousand hectares while average national yield was 1.36 ton/ha. A significant improvement in rice production was recorded in the country between 1986-1990 when output increased to over 2 million tons while average area cultivated, and yield rose to 1,069,200 hectares and 2.09 tons/ha respectively. Throughout the period, rice output and yield increased but in the 1991-1995 periods, while output increased, yield of rice declined. The increased output was traced to expansion in area cultivated. On geographical zone basis, the central zone is the largest producer of rice in Nigeria; accounting

for 44 per cent of the total rice output in 2000. This was followed by the Northwest (29%) while the Southwest was the least (4%). These zones however, differ in terms of their competitive advantage in rice production. It is interesting to note that within a zone, there could be more than one rice ecologies or production systems (Singh *et al.*, 1997). Thus, where two distinct ecologies exist, the zone may have a competitive advantage in the production of rice.

Rice is grown in approximately on 3.7 million hectares of land in Nigeria, covering 10.6 percent of the 35 million hectares of land under cultivation, out of a total arable land area of 70 million hectares. 77 percent of the farmed area of rice is rain-fed, of which 47 percent is lowland and 30 percent upland. The range of grown varieties is diverse and includes both local (such as Dias, Santana, Ashawa, Yarsawaba, and Yarkuwa) and enhanced varieties of traditional African rice such as NERICA (Bayou 2009).

Paddy rice production has been increasing between 2001 and 2006, followed by a decline in 2007 and a positive peak in 2008. From 2008 to 2010 production statistics show a decreasing trend in production, associated with a decline in area harvested between 2006 and 2010. This trend resulted into higher yields between 2008 and 2010, despite declining production. Increasing production between 2002 and 2006 can be explained as result of the implementation of the Presidential Initiative on increased Rice Production, although decreasing production between 2008 and 2010 is not in line with policies aimed at the

development of the rice sector during those years, such as the National Rice Development Strategy and the Federal Market Stabilization Programme (Erenstein 2003).

The main areas of rice cultivation in the country include the middle belt and Northern states of Benue, Kaduna, Niger and Taraba, as well as the South Eastern states of Enugu, Cross River and Ebonyi. Kaduna is the main producing state, followed by Niger, Benue, Ebonyi, Taraba, Kano and Borno. The latter seven states account for over 67 percent of total rice production in the country.

Rice is both a food and a cash crop for farmers, contributing to smallholders' revenues in the main producing areas. WARDA estimates that per capita rice consumption in Nigeria has nearly doubled between the 1980s and 2006, growing from 15.4 kg/year to 25.4 kg/year (WARDA, 2006).

Overall, consumers' preferences have tended to shift away from traditional food towards rice, particularly in urban areas, where rice consumption is increasing the most. The shift in consumer's preferences is due to rising urbanization patterns, population, and income growth, as well as changes in family occupational structures. Changing preferences among consumers are currently influenced by factors such as rice cost, ease, and time of preparation (Odoemena *et al.*, 2008). As a result, rice consumption is particularly growing in cities such as Lagos, Abuja and Makurdi (yearly consumption of 64, 64 and 72 kg/capita, respectively), where most of the urban consumption is met by

imports, while demand is projected to grow 15 percent between 2010 and 2018 (USAID 2009).

Domestic rice is normally semi-milled brown rice, de-hulled, not polished, has great color variation and might contain different varieties in the same bag. In general, there is limited investment in the processing of local rice in Nigeria, and specifically in packaging and improved technology for cleaning paddy and de-stoning (USAID, 2009). Imported Rice is generally processed milled rice. Domestic rice is normally 20-30 percent less expensive than imported rice. The main determinants in price difference between consumed and imported rice are: appearance, the cleanliness, swelling capacity, taste and the homogeneity of the imported rice. However, despite the price and quality differential, there is still an overall acknowledgment of higher organoleptic properties of local rice (Lançonet *al.*, 2003).

Consumers prefer parboiled rice whether imported or local. Nigeria is the main market in West Africa for parboiled rice, while other countries prefer regular milled white rice (USDA FAS, 2005-2010). However, while parboiled rice is particularly appreciated in Nigeria, taste varies across states. For example, non-parboiled rice is preferred in Ekiti state, while parboiled is preferred in Niger state (the second most producing state in Nigeria). Overall, imported rice is preferred to the locally grown rice, based on its superior quality (Diagne, 2011).

2.5.Raw materials for Brewing

2.5.1. Malt and adjuncts

During malting, germination occurs which breaks down cell wall structures and develop enzymes to modify starch granules to soluble starches. The most important contribution from malt in the brewing process is to deliver fermentable sugars and the maltodextrins. The fermentable sugars are the largest part of the malt derived components and will yield ethanol in the final beer. The maltodextrins are not degraded any further by the malt enzymes and are (partly) responsible for creating some 'mouth feel' in the final beer. Apart from these very important ingredients, malt also delivers several other important ingredients such as proteins, lipids, polyphenols and complex Maillard reaction products. The latter are responsible for the typical malt flavor, and for the color (Larlor, 1999). These can be adapted according to the wishes of the brewer, by modifying the last step in the malting process. Proteins can be divided into two groups. Firstly, the non-catalytically active protein delivers a nutritional element to beer, both for the human consumer, but also, when degraded by enzymes to free amino acids and peptides, for the yeast to serve as a nitrogen source during the growth phase in fermentation. Some non-catalytic proteins are responsible for the foaming character of beer and they also contribute to some extent to mouth feel. The second groups of proteins are the enzymes, which are catalytically active. They can convert one component into another.

Polyphenols play a role in the brewing process, as will be discussed later, but also have an impact on colour, mouth feel (astringency) and stability of the final

beer. Lipids play a role during fermentation, as they provide the yeast with membrane material during growth. Otherwise, they may impart off-flavour.

2.5.2 Hops

The hops contribute floral, citrus, herbal aromas, and flavors to beer product. Particularly, soft resin of the hops contains α -acid and β -acid which are the primary bittering compounds (O'Rourke, 2002b). There are three major forms of α -acid, and three analogous forms of β -acid. The α -acids are cohumulone, humulone, and adhumulone, whereas as the β -acids are colupulone, lupulone, and adlupulone. The proportions of these compounds are remarkably consistent on hops varieties affecting on beer flavor and taste.

Several authors have suggested that the effect of antibiotic and positive antibacterial properties in hops is due to the rich sources of flavones glycosides, especially xanthohumol, and others prenylated chalcones, which received much attention as cancer chemopreventive agents (Natarajana *et al.*, 2008). Furthermore, rho- α -acids from hops extract illustrated the anti-inflammatory activity and reduction of cardiovascular disease (Hall *et al.*, 2008).

2.5.3 Yeast

Yeast is a processing aid in brewing. Yeast is a biological catalyst which converts the multiple substrates present in wort to the alcohol containing flavor of beer. Raw materials are being consumed (converted), while yeast is growing and is removed from most commercial beers, or it is at least inactivated

(pasteurized)(Hardwick, 1995). In fact, yeast is a biological 'catalyst', a 'living bag of enzymes', which converts the multiple substrates present in wort to the alcohol-containing, flavor some beer.

2.5.4 Water

Water is normally a source of minerals, which will affect processing and taste, but which normally have a negligible effect on the performance of enzymes, with the exception of amylase. A more important parameter is the pH of the mash. The aim of the brewer is to work optimally with a mashing pH of around 5.5 (± 0.1).

2.5.5 Exogenous enzymes as processing aids

In the brewing process several processing aids, like fining agents and enzymes, have been used for a long time (Mactadden and Clayton, 1998). The main (traditional) reason is that the enzymes in malt are not always present in enough quantities to run the process optimally. Exogenous enzymes, in contrast to endogenous enzymes, are added separately to a process step.

2.6 Beer Production

There are 4 main steps for production of beer and these include: malt production, wort production, beer fermentation, and beer aging.

2.6.1 Malt production

Malting is the first step in beer production, which generates enzyme ingermination of grain kernel and causes certain changes in its chemical constituents. Barley malt is directed by several grain properties such as the

content and composition of proteins, carbohydrates, endosperm structure, cell wall composition and the activities of hydrolytic enzymes during malting (Zhao *et al.*, 2006).

The processes of malt production consist of the following steps;

2.6.1.1 Steeping

Steeping process involves hydrating the grains in order to stimulate the grains for germination. The process is carried out by soaking the grains, draining and air rest. The correct combinations of water and air rest must be observed. At approximately 35% moisture content, the embryo within each kernel of grain is starting to germinate. The modification of rice or barley kernel is satisfactory if they have the appropriate moisture content at the end of steeping (Gamlath *et al.*, 2008).

2.6.1.2 Germination

Germination is a process used to produce malt enzymes for brewing. The germination starts during the air rests towards the end of the steeping stage. A technical term of “modification” is used to describe either all the physical and chemical changes that occur when grains are converted into malt caused by the degradation of the cell walls of starchy endosperm (Briggs, 1998). Gibberellic acid (GA) is synthesized by the embryo and diffused to the aleurone layer cell. Then, the aleurone layer is induced and amylolytic enzymes are

released into the endosperm. Subsequently, the α -amylase and β -amylase convert the starch molecules of the grain into sugars that the embryo can use as food.

2.1.6.3 Kilning

Kilning is the drying process designed for reducing the moisture content in germinated grain from over 43% to less than 5%. Water removal was done by passing hot air through malt and germination of grain should be terminated. The reduction of moisture stabilizes the grain and allows for long-term storage, which inhibits all life processes in malt for example, enzyme activity and modification in grain.

During kilning, there is a development of color and increase in the acceptable flavors (Bamforth, 2006).

Color development results from the reaction between sugar and amino acids at high temperature to form melanoidins by Maillard reaction (Palmer, 2006). After the kilning process, the rootlets are cut off and removed through a machine known as Deculmer to remove the culm or small rootlets that have emerged from each kernel during germination.

2.6.2 Wort production

The purpose of wort production is to provide the necessary conditions for fermentation of sugar from yeast to alcohol, and carbon dioxide. The initial components in malt must be converted to soluble fermentable sugar.

2.6.2.1 Malt milling

To provide malt enzymes, the malt must be broken into small fragments. The husks must be treated carefully and used as a filter material during lautering.

2.6.2.2 Mashing

Mashing is an important process in wort production because, it is the stage where fermentable sugars are produced. Mashing is however, the process of mixing the crushed malts and the cereal adjuncts if used, with water and letting the mixture stand while the enzymes degrade the proteins and starch to yield soluble malt extract called wort. Malt is added to heat with purified water, carefully controlled time, and temperature process. The malt enzymes break down the starch to sugar and non-fermentable sugar (dextrin) as well as the complex proteins of the malt is converted to simpler nitrogen compounds. During mashing, two amylase enzymes including α -amylase and β -amylase present the important function for starch hydrolysis. Alpha-amylase has optimal temperature in range of 70-75°C and β -amylase is around 55-60°C. The protease enzyme can work at temperature 45-50°C and FAN is determined as product of enzyme activity. Carbohydrate composition of various types of wort was showed in the mashing methods can be divided into two main categories: infusion and decoction mashing. The infusion mashing is done at a single temperature without stirring, which requires high-quality and well germinated malt (Eaton, 2006). The decoction method is used as a series of different temperature with under-modified or enzymatically weak malt (Montanari *et al.*, 2005).

2.6.2.3 Lautering

At the end of the mashing process, the mash consists of wort mixture of undissolved substance called “spent grains”. The spent grains consist essentially of the husk, seedling, and other materials, which are the source of protein and fiber that provide the benefits with human (Mussatto *et al.*, 2006). Only wort is used for beer production and spent grains must be separated. The mash is transferred to a lautering vessel usually use cylindrical with a slotted false bottom. The liquid extract drains through the false bottom and run off to the brew kettle. The extract through the grains is washed out as much of the extract as possible. The spent grains are removed and sold as animal feed.

2.6.2.4 Wort boiling and hopping

This serves numerous technological important functions including:

- a) Inactivation of residual enzymes activity after mashing process.
- b) Sterilization of the wort to eliminate all bacteria, yeast and moulds that could participate with the brewing yeast and possibly cause off-flavor.
- c) Coagulation of excess proteins and tannins to form solid particles (trub), which are important for beer stability and foam. The key reaction during protein coagulation related with the destruction of disulfite bridges, which convert to free thiol-groups. These can be reacted with thiol-groups of another proteins and peptides.
- d) Formation of dimethyl sulfite (DMS) from the precursor methylmethionine (SMM) during wort boiling. However, the developments of DMS also appear through kilning and fermentation process. The DMS has a significant impact on beer flavor and is the most significant flavor compound from malt. Typically, the aroma of DMS

is described as a cooked sweetcorn, tinned tomato or baked bean flavor (Bamforth, 2009).

e) Extraction of the bittering compounds from hops.

f) Color formation by Maillard reaction.

2.6.3 Fermentation and biochemistry of fermentation

To transform wort into beer, the sugar in wort must be fermented by yeast to ethanol and carbon dioxide. The consequence in the formation of fermentation and by-product has considerable effect on taste, aroma, and other characteristic properties of beer. For the fermentation of sugar and the carbohydrate metabolisms such as glucose, fructose, maltose, and maltotriose are transported into the cell. The enzyme invertase is responsible for the hydrolysis of sucrose. Glucose and fructose are converted in the cytoplasm and a series of complicated intermediate stages. Pyruvate is produced and ultimately converted to ethanol. The conversion of glucose to 2 pyruvates within 10 intermediate stages is known as Glycolysis. Under the absence of oxygen, yeast converts pyruvate to ethanol through alcoholic fermentation. However, the presence of oxygen fermentation is greatly inhibited or totally stopped called "Pasteur effect" the pyruvate is transported to mitochondria and breakdown via many intermediate stages to achieve high energy (36 ATP/mol), CO₂, and H₂O. Unfortunately, in the classical beer production process, wort aeration step is still necessary to ensure enough yeast growth, and a good fermentation performance. The wort aeration has a negative impact on wort qualities leads to oxidation which, can damage the flavor, and ethanol content (Depraetere *et al.*, 2008)

2.6.4 Aging and finishing

Aging refers to flavor maturation at the end of fermentation, many undesirable flavors, and aroma of “green beer” or “immature beer” is presented. The purpose of aging is to reduce the levels of undesirable compounds including the diacetyl or the sulfur compounds as well as stabilization of beer. The diacetyl or vicinal diketones (VDKs) is the normal products of brewery fermentations that impart to beer characteristic aroma which provided a buttery flavor considered objectionable in lighter-bodied beer, if their concentration is 0.1 – 0.14 mg/L. During aging, yeast produced α -acetolactate then secreted into wort and converted to diacetyl and 2,3 -butanedione.

After aging, clarification step, it is required to remove any remaining yeast as well as suspend particles of protein-polyphenol complexes and insoluble materials during cold storage. The common clarification techniques are used either or combination including sedimentation, fining, centrifugation, and filtration damage the flavor, and ethanol content (Depraetere *et al.*, 2008).

CHAPTER THREE

MATERIALS AND METHODS

3.1. Schematic Overview of Experimental Study

The schematic overview of experimental study, showing the key/major stages from the procurement of rice samples through malting, wort production and beer production to laboratory analyses, is shown in Figure 3.1. Specifically, this current work was designed to evaluate the optimal malting conditions of three

rice varieties for improved brewing trials, as influenced by varying malting conditions. This involved experimental variables, namely: steeping durations (18 h, 24 h and 30 h), germination periods (2, 3 and 4 days) and kilning temperatures (50 °C and 55 °C).

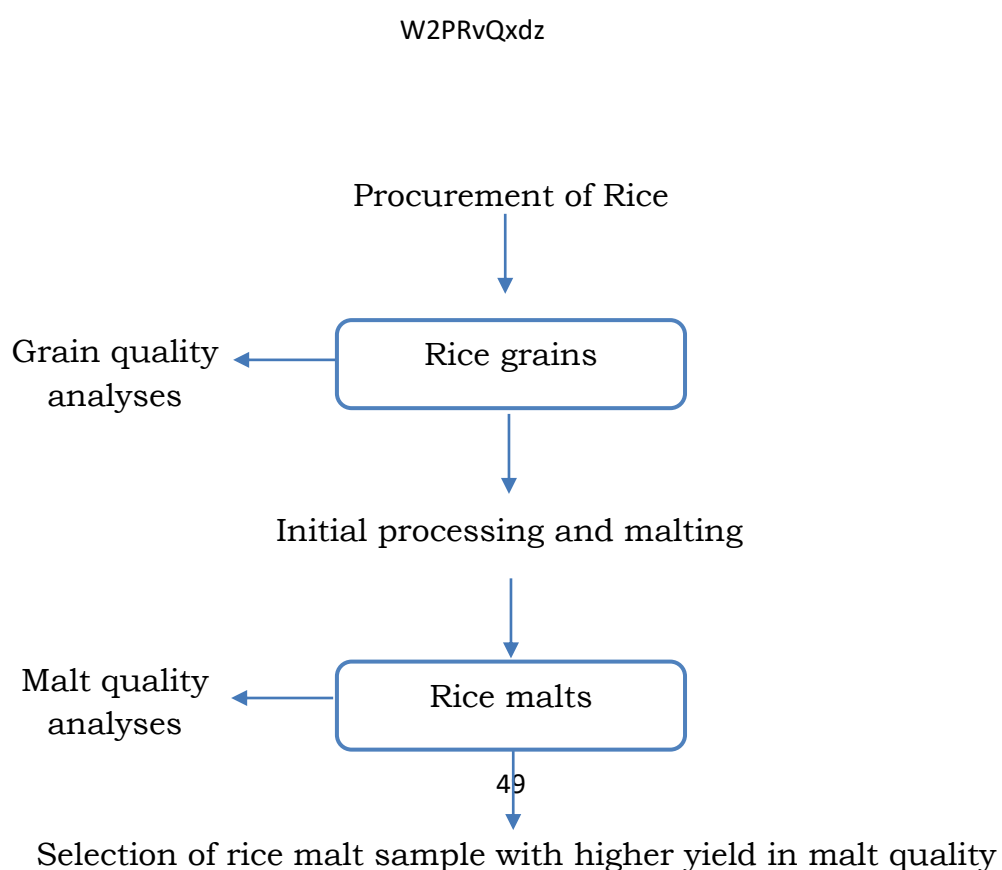


Figure 3.1: Schematic overview of the experimental program.

3.2. Procurement of Chemicals, Enzymes and Rice grains

Procured from certified sources, all chemicals and reagents (i.e., Copper (II) sulphate pentahydrate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$), Potassium sodium tartrate tetrahydrate ($\text{KNaC}_4\text{H}_4\text{O}_6 \cdot 4\text{H}_2\text{O}$), Calcium hydroxide ($\text{Ca}(\text{OH})_2$), hydrochloric acid (HCl), boric acid (H_3BO_3), potassium sulphate (K_2SO_4), sulfuric acid (H_2SO_4), Trioxonitrate (V) acid (HNO_3), sodium metabisulphite ($\text{Na}_2\text{S}_2\text{O}_5$), Sodium

hydroxide (NaOH), Ammonium hydroxide (NH₄OH), Ninhydrin, Methylene blue indicator, Phenolphthalein indicator, Fehling's solution, Anhydrous D-glucose) were of analytical grade standard.

Commercial exogenous enzymes (namely: α -amylase and β -amylase) were procured from Nigerian breweries Plc (Awo-Omamma, Imo State, Nigeria). Protease enzyme and isomerised hops used were procured from Department of Applied Microbiology and Brewing, Enugu State University of Science and Technology, Enugu State, Nigeria. Yeast strain (*Saccharomyces pastorianus*) was procured from the Nigerian Breweries Plc (Ama, Enugu State, Nigeria).

Improved rice grains (FARO 44, FARO 57, and NERICA 7) were purchased from National Cereals Research Institute, Amakama, Olokoro Umuahia, Abia State, Nigeria.

3.3. Processing of Rice Samples

3.3.1. Malting of Rice

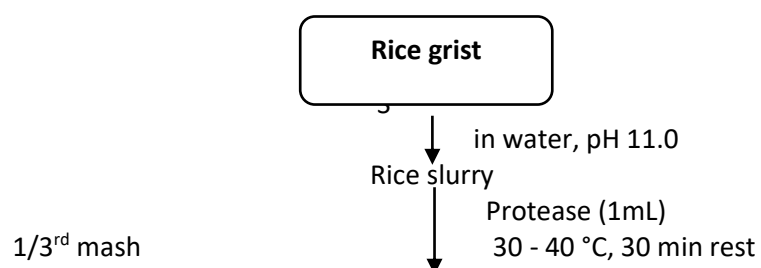
After manual cleaning, the paddy rice of different varieties were sorted to remove contaminants/damaged seeds and was winnowed to remove dust. Prior to malting, the rice paddy was disinfected in water containing 0.20 % sodium metabisulphite. The malting process followed method of Kunze (2005) with some modifications. Briefly, rice samples were steeped in water at 20 - 25 °C

for 18, 24 and 30 h with alternating steep cycle of 6 h wet-steep period and 30 min air rest. Grains were allowed to germinate, thereafter removed after 2, 3 and 4 days and kilned in hot air oven (Genlab, England, Model M 30 C, S/N 92B060) at temperatures between 50 – 55 °C for about 22 – 24 h. Kilned samples were manually de-rooted by rubbing off with hand, winnowed to remove the rootlets/dust and milled to produce the rice grist (see figure 3.1).

3.3.2. Production of Rice Malt Wort

The flow diagram of rice malt wort production, presented in Figure 2, slightly modified from the previously studies (Nwanekezi,*et al.*,2007; Ofoedu *et al.*, 2019; Iwouno,*et al.*, 2019), which involved a three-step decoction mashing process. Rice malt grist (~ 2 kg) was dissolved in clean filtered potable water (8 L) previously made to a pH of 11.0 using Ca(OH)₂ solution. The entire mash temperature was raised to 35 – 40 °C to acidulate the mash followed by addition of 1 mL of protease for proteolysis to take place, with temperature maintained for 30 min at gentle stirring (acid rest). First decoction involved transferring one-third of the mash to a mash kettle and heated to 70°C. Heated mash was transferred back to the remaining two-third mash, with entire temperature raised to 50 – 55°C, followed by addition of α-amylase (0.8 mL) and subsequently allowed to rest for 30 min (protein rest). Second decoction involved one-third of the mash was further heated in the mash kettle (3 - 5 min) until temperature of 85°C was reached, then transferred to the remaining two-third thin mash, raising the temperature to approximately 67°C approximately and the mash

gelatinized, and allowed to rest for 30 min after addition of α -amylase (0.8 mL). Following liquefaction, a third decoction involved raising temperature to 100°C, wherein boiled mash was added to the remaining mash, which moderated the entire temperature to about 72 – 75°C. Mash rested again for approximately 30 min after addition of β -amylase (0.8 mL) prior to saccharification. Iodine test was carried out to determine the extent of degradation of starch. To denature the enzymes prior to wort lautering, mashing-out was carried out. Spent grains were sparged with 1000 mL of hot sparge water at 80 °C to obtain the entire wort from the mashing operation, before wort was concentrated.



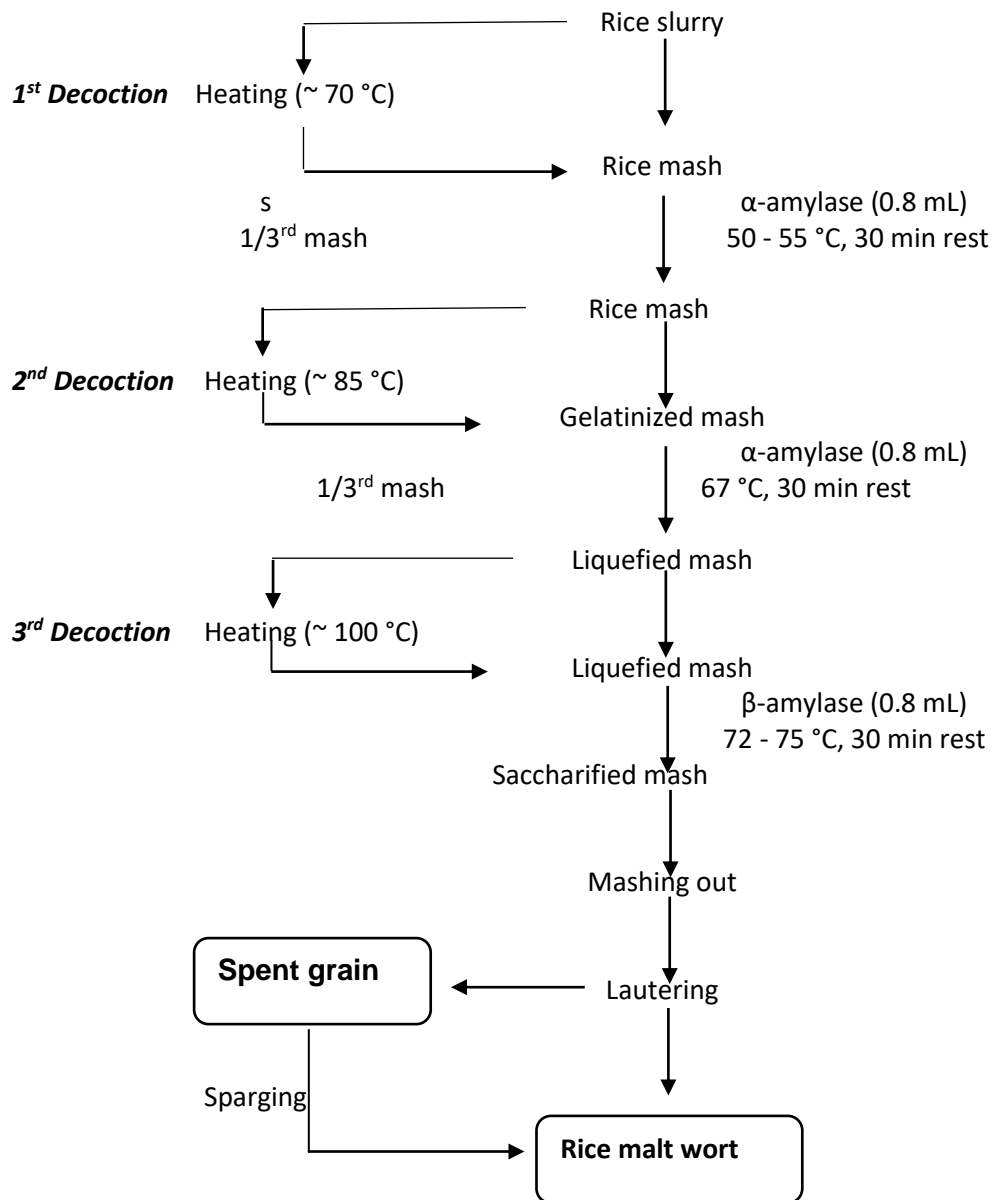


Figure 3.2: Flow diagram to produce rice malt wort

3.3.3. Production of Rice Malt Beer.

Adopting the Japanese rice lager production process, however, the rice malt beer in this study was produced according to the method described by Briggs *et*

al. (2004) with slight modification. Briefly, rice malt wort was boiled together with hop extracts for 30 min and thereafter, allowed to cool. Undissolved particles were removed and the filtered wort was transferred into fermenters. Already activated yeast (3g) culture (*Saccharomyces pastorianus*) was pitched into the fermenting vessel at 20 °C and fermentation was carried out at 10 – 20 °C for 8 days. Green beer was filtered and allowed to age for 21 days. Matured/draft beer was siphoned into sterile bottles, pasteurized at 60°C for 15 min, and subsequently, analyzed.

3.4. Evaluation of Rice Samples.

this current work, thousand corn weight, germinative energy, germinative capacity, and degree of steeping were determined in accordance to a method described by (Marconi, *et al.*, 2017)..

3.4.1. Determination of Thousand Corn Weight (TCW)

This was determined according to the method described by Esiape (1994). Hundred (100) grains of paddy rice randomly selected from the bulk were weighed using a weighing balance. Each weight was multiplied by 10 to obtain the 1000 kernel weight. Determinations were done in triplicate.

3.4.2. Determination of Germinative Capacity (GC) and Germinative Energy (GE)

Germination capacity measures grain viability, whereas Germination capacity measures the extent to which grain will germinate in a standardized test. Both

rapid and complete germination are well-known essential features of good malt. For this current work, the GE and GC (presented in percent [%]) of rice samples was determined using the recommended method of analysis of the Institute of Brewing (IoB) (2007).

$$\text{Germinative energy (\%)} = \frac{\text{Number of viable grains}}{\text{Total number of grains}} \times 100 \quad \text{Eq.3.1}$$

$$\text{Germinative capacity (\%)} = \% \text{ Germinative energy} - \% \text{ Dormancy} \quad \text{Eq. 3.2}$$

Herein, the dormancy is rice grain's inherent inability to germinate under optimal environmental conditions expressed as;

$$\% \text{ Dormancy} = \frac{\text{Number of unviable grains}}{\text{Total number of grains}} \times 100 \quad \text{Eq. 3.3}$$

3.4.3. Degree of steeping (DoS)

DoS measures the amount of water readily absorbed by the grains. DoS expressed as percentage (%) was determined by the method described by Kunze (2004) with slight modification. One hundred rice kernels of each variety, the moisture content of which has been previously determined were soaked in a 100 ml beaker containing 50 ml of distilled water at ambient temperature ($28^{\circ}\text{C} \pm 2$). Steeping was done until constant weights were attained and recorded. Soak waters were drained off the grains using sieves. From the increase in mass, the degree of steeping was calculated as;

$$\text{Degree of steeping (\%)} = \frac{X}{W_i} \times 100 \quad \text{Eq. 3.4}$$

$$\text{Where } X = \frac{W_i \times (D + MC)}{W_f}$$

W_i = Mass of rice grain before steeping

W_f = Mass of rice grain after steeping

MC = Moisture content of rice grain

D = Mass of water absorbed by rice grain *(W_f – W_i) (g)

3.5. Characteristic Analyses of Rice Malt, Wort and Beer samples

3.5.1. Rice Malt Analyses

Malt analyses are carried out in accordance with standard methods of the Institute of Brewing (IoB), European Brewing Convention (EBC) and American Society of Brewing Chemists (ASBC) for several purposes such as to provide data for the maltster to use for quality control and to guide process adjustments, to provide a basis for product valuation, for prediction of extract recovery, to indicate the potential value of the malt and whether or not a particular malt is likely to give production difficulties (Briggs, 1998).

3.5.1.1: Determination of Cold Water Extract(CWE)

Cold Water Extract (presented in g/100g) was determined using method recommended by Institute of Brewing (IoB) (2007) and was calculated using equation below:

$$\text{Cold Water Extract (\%)} = \frac{(G - 100)}{3.86} \times 20 \quad \text{Eq. 3.5}$$

where,

G = the excess degrees of gravity of the filtrate at 15.5 °C as 1000

i.e. G = 1000 (S.G - 1)

3.4.1.2. Determination of Hot Water Extract (HWE)

HWE measures the extractable materials derived from malt after a small scale mashing process (Briggs, 1998). HWE expressed as liters degrees/kg (°L/kg) was determined using method recommended by Institute of Brewing (IoB) (2007) and was calculated using the equation below;

$$\text{Hot Water Extract (°L/kg)} = \frac{G \times V}{M} \quad \text{Eq.3.6}$$

where,

G = the excess degrees of gravity of the filtrate at 15.5 °C as 1000

i.e. G = 1000 (S.G - 1)

V = Volume of wort in Litres (L)

M = Mass of malt in Kilogram (kg)

3.5.1.3. Determination of Diastatic Power (DP)

DP measures the amount of enzyme in malt available to convert complex carbohydrates/starches into fermentable sugars (Ackley, 2018). DP expressed in lintner degree (°L) was determined using method recommended by Institute of Brewing (IoB) (2007) and was calculated using the equation below;

$$\text{DP} = \frac{(2000 - 200)}{(xy - xs)} \quad \text{Eq.3.7}$$

where,

x = mL of malt extract

y = mL of converted starch to 5 mL of the Fehling's solution

s = Titre for starch blank

3.5.1.4. Determination of Moisture Content (MC) and Total Nitrogen (TN)

Malt with higher diastatic power (DP), cold water extract (CWE) and hot water extract (HWE) were selected for wort production/brewing trial. Only these were subject to determinations of moisture content (MC) presented in g/100g (wet basis) via the method described in AOAC (2006), and total nitrogen (TN) presented in g/L via the Kjeldahl method (European Brewery Convention, 2006).

3.5.1.5. Malting Loss (ML)

Malting loss (ML) after germination was determined according to the method described by Adebowale *et al.*, (2010) by weighing the rice grains before and after malting. The weight of 100 grains of rice was recorded before malting and the weight of the malted grains after the rootlets were removed by hand was also recorded. Malting loss was expressed as percentage (%) on dry matter basis.

$$\text{Malting loss (\%)} = \frac{\text{Weight of unmalted grain} - \text{Weight of malted grain}}{\text{Weight of unmalted grain}} \times 100 \quad \text{Eq. 3.8}$$

3.5.1.6. Malt Yield (MY)

Malt yield (MY) after germination was determined according to the method described by Adebowale *et al.* (2010) by weighing the rice grains before and after malting. The weight of 100 grains of rice was recorded before malting and the weight of the malted grains after the rootlets were removed by hand was

also recorded. Malt yields were expressed as percentage (%) on dry matter basis.

$$\text{Malt yield (\%)} = \frac{\text{Weight of malted grain}}{\text{Weight of unmalted grain}} \times 100 \quad \text{Eq. 3.9}$$

3.5.2. Rice Wort Analyses

3.5.2.1. Determination of pH

pH of rice malt wort was determined using the method described by AOAC (2004). Digital pH meter calibration used buffer 4, 7 and 9 solutions at 25 °C. pH conducted measurement required electrode (probe) dipping into each 25 mL pipetted wort sample, allowed to stabilize before reading off.

3.5.2.2. Determination of total soluble nitrogen (TSN)

TSN measures the nitrogen materials (amino acids, peptides and polypeptides) solubilized by proteolysis during malting and extracted during mashing (Agu & Palmer, 1998; Noonan, 2003). TSN of rice malt wort, expressed in g/L was determined using Kjeldhal method (Institute of Brewing (IoB), 2007).

3.5.2.3. Determination of Apparent Brix (°Bx)

Brix of rice malt wort was determined using a Milwaukee Digital brix Refractometer Model MA871 (Milwaukee Instruments, NC - USA) (Montañez-Soto *et al.*, 2013), which involved refractometer standardized with distilled water at 20 °C until brix value read zero, followed by two drops of wort sample on the lens (sensitive surface), and measurement conducted subsequently.

3.5.2.4. Determination of Kolbach Index (KI)

KI measures the degree/extent of protein modification/degradation, as a ratio of Total Soluble Nitrogen in wort to Total Nitrogen in the malt (Bamforth, 2003; Olivier & Colicchio, 2012). KI expressed in %, was calculated consistent with Analytical - EBC (1998) method, using the equation below:

$$KI = \left(\frac{TSN}{TN} \right) \times 100 \quad \text{Eq 3.10}$$

3.5.2.5. Determination of Free Amino Nitrogen (FAN)

FAN of rice malt wort was determined by Ninhydrin method (European Brewery Convention, 1998) with slight modifications. The sample (1mL) diluted with deionised water to 100 mL, then 2 mL of diluted sample mixed with 1 mL of colour reagent, placed in boiled water for 16 min, allowed to cool to 20 °C. Diluted solution (5 mL) was added, followed by measurement of optical density at 570 nm. Blank was determined with 2 mL of deionized water. Glycine standard solution was checked using 2 mL of glycine solution. The FAN content was calculated and expressed in mg/L.

3.5.2.6. Determination of Dextrose Equivalence (DE)

DE measures the amount of reducing sugars present in a sugar product, relative to glucose (Dziedzic and Kearsley, 1995) determined on rice malt wort, using the Lane and Eynon Fehling's solution method as previously described (International Starch Institute, 1999).

3.5.2.7. Determination of Original Extract (OE)

OE measures wort density compared to that of water at equal volume and temperature (ASBC, 2009). OE of rice malt wort presented in g/100g was calculated from an approximate Plato value as previously described (Kunze, 2004), calculated using equation below:

$$\text{Specific Gravity (SG)} = (\text{Mass percent} \times 0.004) + 1 \quad \text{Eq.3.11}$$

where Mass percent \equiv Apparent brix value

$$\text{Original Extract (g/100g)} = 259 - \left(\frac{259}{\text{SG}} \right) \quad \text{Eq.3.12}$$

3.5.2.8. Determination of Wort Sugar Profile

The sugar profile of rice malt wort was determined using HPLC according to the method described by AOAC (2006).

3.5.3. Rice Malt Beer Analyses

Rice malt beer was characterized using the following Analytical-EBC methods.(European Brewery Convention, 2007).

3.5.3.1. Determination of pH, Colour, and Apparent Extract.

pH was determined using the EBC method 9.35, similar to AOAC (2004). Colour presented in °EBC was determined using the Spectrophotometric Method (EBC method 9.6). Apparent extract (AE) presented in g/100g was determined using the EBC method 9.43.1.

3.5.3.2. Determination of Alcohol and Turbidity.

Alcohol by volume (%ABV) was determined by distillation (EBC method 9.2.1). Turbidity presented in NTU (Nephelometric Turbidity Unit) was determined by EBC method 9.10.

3.5.3.3. Determination of Sensory Attributes.

A hedonic scale test was used to evaluate the sensory attributes of beer samples according to the method reported by Iwe (2002). This specifically involved comparing the rice malt beer samples with the commercial lager beer. The sensory evaluation was carried out by 20 semi-trained panelists of different age groups (20–36 years old). The prerequisites for participating in the study were that the individual consumed beer and showed interest in participating in all test sessions. Importantly, the participation in this sensory evaluation was voluntary, and oral consent was obtained prior to participation. The coded rice malt beer samples were randomly served at temperatures of about 10°C. Participants were served with 4 series of beer samples in transparent glass cups and the degree of liking was rated using a nine-point hedonic scale with the ratings of 9 as liked extremely and 1 as disliked extremely for five main attributes i.e., colour, aroma, taste, mouthfeel and appearance; while overall acceptance of the samples was evaluated by taking the average of other attributes. The panelists drank potable water to rinse/clean their mouth between tastings to avoid cross-contamination between samples. After tasting, score sheets were filled by the tasters.

3.6. Statistical Analysis

One way analysis of variance (ANOVA) was carried out using IBM SPSS version 20 Software (IBM, New York, USA). Results, of duplicate measurements, were considered statistically significant at $p < 0.05$, and expressed as mean \pm standard deviation (SD). Mean differences were resolved using least significant differences (LSD) at post-hoc conditions. Where required, simple correlation was performed and R-square values reported

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 RESULTS

The grain characteristics of rice varieties is shown in Table 4.1. Specifically, the grain characteristics were depicted in terms of Thousand corn weight (TCW[g]), germinative energy (GE [%]), germinative capacity (GC [%]), and degree of steeping (DoS [%]). Results showed that TCW, GE, GC, and DoS varied significantly ($p < 0.05$) across the rice varieties. Further, the TCW ranged between 25.81 – 27.01 g, GE ranged between 86.50 – 95.00 %, GC ranged between 92.50 – 95.50 %, and DoS ranged between 49.96 – 55.00 %. Clearly, the TCW peaked at NERICA 7 (27.01 g). Additionally, both GE and GC peaked at NERICA 7, with 95.00 %, and 95.50% respectively. Additionally, the DoS was peak (55.00 %) at FARO 44, and least at NERICA 7 (49.96 %). Besides, the rice varieties obtained acceptable grain germination of above 85 %.

The cold water extract (CWE [g/100g]), hot water extract (HWE [L°/kg]) and diastatic power (DP [°Lintner/kg]) of rice malts subject to varying steeping durations, germination periods, and kilning temperatures are shown in Tables 4.2, 4.3, and 4.4, respectively. Results showed that CWE, HWE, and DP in rice malt varied significantly ($p < 0.05$) across the varieties. Specifically, the CWE ranged between 10.95 – 23.16 g/100g, HWE ranged between 51.06 – 206.48 L°/kg, and DP ranged between 22 – 150 °Lintner/kg, across rice varieties. With respect to S₁₈G₃K₅₅^o (18 h steeping, 3 days germination, 55°C kilning),

S₃₀G₂K₅₀^o (30 h steeping, 2 days germination, 50°C kilning) and S₂₄G₃K₅₅^o (24 h steeping, 3 days germination, 55°C kilning) combinations, the CWE, HWE, and DP obtained peaks at FARO 44 (22.88 g/100g; 136.56 L^o/kg; 150 °Lintner/kg), FARO 57 (22.13 g/100g; 170.18 L^o/kg; 148 °Lintner/kg), and NERICA 7 (23.16g/100g; 206.48 L^o/kg; 143 °Lintner/kg). Based on these CWE/HWE/DP peaks, the SGK (steeping duration, germination periods and kilning temperatures) combinations of rice varieties trended as follows: CWE = NERICA 7 (23.16g/100g) > FARO 44 (22.88 g/100g) > FARO 57 (22.13 g/100g); HWE = NERICA 7 (206.48 L^o/kg) > FARO 57 (170.18 L^o/k) > FARO 44. (136.56 L^o/kg); and DP = FARO 44 (150 °Lintner/kg) > FARO 57 (148 °Lintner/kg) > NERICA 7 (143 °Lintner/kg). Outside this specific SGK combinations, the CWE/HWE/DP obtained no peaks.

The malt yield (MY), malting loss (ML), moisture content (MC), and total nitrogen (TN) of rice malt samples is shown in Table 4.5. Results showed that ML and MY of rice malt changed significantly ($p < 0.05$) across the varieties. The MY ranged between 87.11 – 92.65 %, whereas the ML ranged between 6.03 – 10.80 %. Additionally, the peak (10.80 %) ML and least (87.11 %) MY can be seen at NERICA 7, whereas the peak MY (92.65 %) and least ML (6.03 %) can be seen at FARO 44. Specifically from FARO 44 (S₁₈G₃K₅₅^o), FARO 57 (S₃₀G₂K₅₀^o) and NERICA 7 (S₂₄G₃K₅₅^o), the MC and TN of rice malts were determined (also shown in Table 4.5). Results showed that both MC and TN

varied significantly ($p < 0.05$). Specifically, the MC of rice malts ranged between 5.19 – 6.43g/100g, whereas the TN of rice malts ranged between 13.10 – 15.70g/L. Additionally, the MC in rice varieties trended as follows: NERICA7 (6.43g/100g) > FARO44 (5.52g/100g) > FARO57 (5.19g/100g), whereas the TN in rice varieties trended as follows: FARO 44 (15.70g/L) > FARO 57 (14.30g/L) > NERICA 7 (13.10g/L). Additionally, the peaks of MC and TN were obtained at NERICA7 (6.43 ± 0.14 g/100g) and FARO44 (15.70 ± 0.05 g/L), respectively.

The sugar profile of rice malt wort samples is shown in Table 4.6. A combination of sugars (glucose, maltose, maltotetraose, maltotriose, raffinose, and sucrose) can be seen, which significantly differed ($p < 0.05$) across varieties. In particular, the glucose ranged between 10.84 – 11.63 %, maltose ranged between 14.63 – 15.34 %, maltotetraose ranged between 0.44 – 0.63 %, maltotriose ranged between 12.26 – 16.40 %, raffinose ranged between 0.05 – 0.07 %, and sucrose ranged between 2.32 – 2.83 %. Additionally, the sugars in rice malt wort trended by varieties as follows: glucose = FARO57 > FARO44 > NERICA7; maltose = FARO57 > NERICA7 > FARO 44; maltotetraose = FARO44 > FARO57 > NERICA7; maltotriose = NERICA7 > FARO57 > FARO44; raffinose = FARO44 > NERICA7 > FARO57; and sucrose = NERICA7 > FARO44 > FARO57. From these, we see that FARO57 obtained peaks at glucose (11.63 ± 0.71 %) and maltose (15.34 ± 0.08 %), FARO44 obtained peaks at maltotetraose (0.63 ± 0.04 %) and raffinose (0.07 ± 0.00 %),

NERICA7 obtained peaks at maltotriose (16.40 ± 0.07 %) and sucrose (2.83 ± 0.08 %).

The pH, total soluble nitrogen (TSN [g/L]), Brix (g/100g), kolbach index (KI [%]), free amino nitrogen (FAN [mg/L]), dextrose equivalence (DE [g/100g]), and original extract (OE [g/100g]) components of rice malt wort samples is shown in Table 4.7. Results showed that, whereas Brix, KI and FAN varied significantly ($P < 0.05$), the pH, TSN, DE, and OE resembled ($p > 0.05$), with the following ranges across rice varieties: pH ranged between 5.30 - 5.40, TSN ranged between 5.40 - 5.80 g/L, Brix ranged between 13.88 - 16.36 g/100g, KI ranged between 34.39 - 44.27 %, FAN ranged between 108.56 - 117.34 mg/L, DE ranged between 37.00 - 40.00 g/100g, and OE ranged between 9.68 - 12.66 g/100g. By rice varieties, therefore, these (above-mentioned) parameters in rice malt wort obtained peaks at FARO44 (pH = 5.40 ± 0.38), NERIA7 (TSN = 5.80 ± 0.00 g/L), FARO57 (Brix = 16.36 ± 0.42 g/100g), NERICA7 (KI = 44.27 ± 0.28 %), NERICA7 (FAN = 117.34 ± 0.06 mg/L), and FARO57 (DE = 40.00 ± 0.82 g/100g) / (OE = 12.66 ± 0.04 g/100g). Additionally, these parameters in rice malt wort trended as follows: pH = FARO 44 (5.40) > FARO57/NERICA 7 (5.30); TSN = NERICA 7 (5.80g/L) > FARO 57 (5.60g/L) > FARO 44 (5.40g/L); Brix = FARO 57 (16.36g/100g) > FARO 44 (14.65g/100g) > NERICA 7 (13.88g/100g); KI = NERICA 7 (44.27%) > FARO 57 (39.16%) > FARO 44 (34.39%); FAN = NERICA 7 (117.34mg/L) > FARO 57 (112.23mg/L) > FARO 44 (108.56mg/L); DE = FARO 57 (40g/100g) >

FARO 44 (39g/100g) > NERICA 7 (37g/100g); and OE = FARO 57 (12.66g/100g) > FARO 44 (11.15 g/100g) > NERICA 7 (9.68g/100g).

The pH, colour (°EBC), apparent extract (AE [g/100g]), alcohol content (%ABV) and turbidity (NTU) of rice malt beer samples is shown in Table 4.8. Results showed that, whereas the alcohol content varied significantly ($P < 0.05$), the pH, colour, AE and turbidity resembled ($p > 0.05$), with the following ranges across rice varieties: pH ranged between 3.80 - 3.90, colour ranged between 3.20 - 3.73 °EBC, AE ranged between 4.57 - 4.93 g/100g, alcohol content ranged between 2.82 - 4.13 %ABV and turbidity ranged between 4.30 - 4.80 NTU. By rice varieties, therefore, these (above-mentioned) parameters in rice malt beer obtained peaks at FARO57 (pH = 3.90 ± 0.59 ; colour = 3.73 ± 0.71 °EBC; and alcohol content = 4.13 ± 0.18 %ABV), NERICA7 (AE = 4.93 ± 0.54 g/100g) , and FARO44 (turbidity = 5.30 ± 0.01 NTU). Additionally, these parameters in of rice malt beer trended as follows: pH = FARO 57 (3.90) > FARO 44 & NERICA 7 (3.80); colour = FARO 57 (3.73 °EBC) > FARO 44 (3.70 °EBC) > NERICA 7 (3.20 °EBC); AE = NERICA 7 (4.93 g/100g) > FARO 44 (4.59 g/100g) > FARO 57 (4.57 g/100g) ; alcohol content = FARO 57 (4.13%ABV) > FARO 44 (3.54 %ABV) > NERICA 7 (2.82 %ABV) ; and turbidity = FARO 44= (5.30NTU) > FARO 57 (4.80NTU) > NERICA 7 (4.30NTU).

The sensory attributes (colour, aroma, taste, mouthfeel, appearance, and overall acceptability) of rice malt beer samples is shown in Table 4.9. Importantly, the sensory attributes of rice malt beer were compared with the commercial lager beer. Results showed that colour, taste, mouthfeel, appearance, and overall acceptability of rice malt beer were significantly ($p < 0.05$) less than those of commercial lager beer. Only the aroma of rice malt beer resembled ($p > 0.05$) that of commercial lager beer. Specific to the rice malt beer, the sensory attributes ranged as follows: the colour ranged between 6.66 – 6.91; aroma ranged between 7.54 – 7.81; mouthfeel ranged between 6.57 – 6.96; appearance ranged between 6.24 – 6.52; taste ranged between 7.69 – 7.87, and overall acceptability ranged between 6.94 – 7.21. By rice varieties with respect to this (rice) malt beer, only the sensory attributes of FARO44 obtained peaks, namely: colour = 6.91; taste = 7.87; aroma = 7.81; mouthfeel = 6.96; appearance = 6.52; and overall acceptability = 7.21.

Table 4.1: Malting characteristics and grain quality properties of rice varieties.

SAMPLES	Thousand Corn Weight (g)	Germination Energy (%)	Germination Capacity (%)	Degree of Steeping(%)
FARO 44	25.81 ^c ±0.01	86.50 ^c ±0.06	92.50 ^b ±0.14	55.00 ^a ±0.16
FARO 57	26.11 ^b ±0.16	93.50 ^b ±0.07	93.00 ^b ±0.00	50.98 ^b ±0.07
NERICA 7	27.01 ^a ±0.07	95.00 ^a ±0.98	95.50 ^a ±0.10	49.96 ^c ±0.09
LSD	0.24	1.50	1.50	0.44

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each parameter is not significantly different (P>0.05)

TCW = Thousand corn weight; GE = Germination energy; Germinative capacity; MY = Malt yield; ML = Malting loss; DoS = Degree of steeping

Table 4.2: Cold water extract (CWE) (g/100g) of rice malts subject to varying steeping durations, germination periods and kilning temperatures

Steeping duration (hours)	Malting conditions		Rice varieties		
	Germination period (days)	Kilning temperature (°C)	g/100g		
			FARO 44	FARO 57	NERICA 7
18	2	50	21.66 ^d ±0.03	19.43 ^c ±0.28	10.95 ^l ±0.01
		55	22.28 ^b ±0.06	14.39 ^j ±0.42	12.68 ⁱ ±0.04
	3	50	21.84 ^c ±0.10	13.99 ^k ±0.06	11.73 ^k ±0.06
		55	22.88 ^a ±0.13	11.44 ^m ±0.06	13.31 ^f ±0.07
	4	50	20.45 ^j ±0.08	16.51 ^h ±0.07	12.68 ⁱ ±0.06
		55	20.53 ^{hi} ±0.17	14.38 ^g ±0.11	20.25 ^c ±0.17
24	2	50	20.77 ^f ±0.14	20.58 ^b ±0.14	12.52 ^j ±0.08
		55	20.95 ^e ±0.14	15.17 ⁱ ±0.10	14.94 ^d ±0.10
	3	50	20.58 ^{gh} ±0.11	18.24 ^e ±0.10	12.52 ^j ±0.07
		55	20.76 ^f ±0.06	13.20 ^l ±0.04	23.16 ^a ±0.07
	4	50	19.81 ^l ±0.08	16.91 ^g ±0.07	12.93 ^h ±0.57
		55	20.49 ^{ij} ±0.08	17.18 ^f ±0.07	21.13 ^b ±0.08
	2	50	18.50 ⁿ ±0.04	22.13 ^a ±0.06	13.32 ^f ±0.03
		55	19.72 ^m ±0.13	16.51 ^h ±0.11	17.19 ^g ±0.11

30	3	50	19.61 ^k ±0.14	18.98 ^d ±0.13	13.50 ^f ±0.10
		55	20.58 ^g ±0.03	15.17 ⁱ ±0.03	14.54 ^d ±0.01
	4	50	18.49 ⁿ ±0.06	19.03 ^d ±0.06	13.69 ^f ±0.04
		55	20.46 ^{ij} ±0.08	16.51 ^h ±0.07	16.34 ^f ±0.07
LSD			0.27	0.59	0.45

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each rice variety is not significantly different (P>0.05)

Table 4.3: Hot water extract (HWE) (°L/kg) of rice malts subject to varying steeping durations, germination periods and kilning temperatures

Malting conditions			Rice varieties		
Steeping duration (h)	Germination period (days)	Kilning temperature (°C)	HWE (°L/kg)		
			FARO 44	FARO 57	NERICA 7
18	24	50	121.56 ^b ±0.48	103.05 ^h ±0.41	130.06 ^h ±0.54
		55	103.18 ^e ±1.02	103.18 ^{gh} ±0.99	182.10 ^b ±1.80
	3	50	103.18 ^e ±0.74	105.17 ^f ±0.75	163.1 ^e ±1.16
		55	136.56 ^a ±1.16	100.15 ⁱ ±0.88	182.10 ^b ±1.54
	4	50	106.22 ^d ±0.59	160.43 ^c ±0.91	121.41 ^k ±0.69
		55	115.56 ^c ±0.81	105.17 ^f ±0.74	163.90 ^e ±1.13
24	2	50	100.15 ^f ±1.13	106.22 ^e ±1.20	166.91 ^d ±1.90
		55	84.97 ^j ±0.83	106.22 ^e ±1.02	148.00 ^f ±1.47
	3	50	100.15 ^f ±0.17	106.22 ^e ±0.18	182.10 ^b ±0.31
		55	91.04 ⁱ ±0.44	103.18 ^{gh} ±0.51	206.48 ^a ±0.99
	4	50	84.97 ^j ±0.54	106.22 ^e ±0.68	182.10 ^b ±1.16
		55	106.18 ^d ±0.28	103.48 ^g ±0.28	133.70 ⁱ ±0.35
2	50	51.06 ^m ±0.33	170.18 ^a ±1.10	139.60 ^g ±0.91	

		55	75.98 ^k ±0.24	136.56 ^d ±0.34	127.50 ^j ±0.41
30	3	50	75.98 ^k ±0.65	166.91 ^b ±1.41	127.50 ^j ±1.08
		55	97.06 ^g ±0.55	106.22 ^e ±0.78	163.90 ^e ±0.93
	4	50	60.70 ^l ±0.21	106.02 ^e ±0.35	172.98 ^c ±0.59
		55	94.21 ^h ±0.62	103.08 ^h ±0.68	133.70 ⁱ ±0.89
LSD			0.49	0.30	1.19

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each rice variety is not significantly different (P>0.05)

Table 4.4: Diastatic power (DP) (°Linter/kg) of rice malts subject to varying steeping durations, germination periods and kilning temperatures

Malting conditions			Rice varieties		
Steeping duration (h)	Germination period (days)	Kilning temperature (°C)	°Linter/kg		
			FARO 44	FARO 57	NERICA 7
18	2	50	78.00 ^e ±0.74	63.00 ^f ±0.59	71.00 ^j ±0.66
		55	92.00 ^c ±0.59	22.00 ⁿ ±0.14	97.00 ^g ±0.61
	3	50	63.00 ^g ±0.28	50.00 ⁱ ±0.23	48.00 ^m ±0.21
		55	150.00 ^a ±0.23	38.00 ^l ±0.06	60.00 ^l ±0.10
	4	50	80.00 ^d ±0.37	37.00 ^l ±0.17	67.00 ^k ±0.30
		55	98.00 ^b ±0.42	26.00 ^m ±0.11	100.00 ^e ±0.44
24	2	50	43.00 ^l ±0.06	80.00 ^e ±0.11	98.00 ^f ±0.14
		55	63.00 ^f ±0.18	39.00 ^k ±0.11	120.00 ^c ±0.34
	3	50	50.00 ⁱ ±0.21	64.00 ^f ±0.27	60.00 ^l ±0.25
		55	52.00 ⁱ ±0.28	52.00 ^h ±0.28	143.00 ^a ±0.79
	4	50	44.00 ^k ±0.34	57.00 ^g ±0.44	60.00 ^l ±0.45
		55	57.00 ^h ±0.61	39.01 ^k ±0.41	101.00 ^e ±1.08
	2	50	38.00 ^p ±0.15	148.00 ^a ±0.58	102.00 ^e ±0.41

		55	39.00 ^o ±0.38	43.00 ⁱ ±0.42	113.00 ^d ±1.12
30	3	50	35.00 ^q ±0.25	135.00 ^b ±0.96	92.00 ^h ±0.65
		55	38.00 ^p ±0.32	98.08 ^c ±0.83	138.00 ^b ±1.17
	4	50	39.00 ^o ±0.23	92.00 ^d ±0.52	46.00 ^m ±0.25
		55	40.00 ^m ±0.24	43.00 ⁱ ±0.25	75.00 ⁱ ±0.45
LSD			0.73	0.98	1.63

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each rice variety is not significantly different (P>0.05)

Table 4.5: Measured values of analyzed rice malt samples

Samples	Malting Yield (%)	Malting Loss (%)	Moisture Content (g/100g)	Total Nitrogen (g/L)
FARO 44	92.65 ^a ±0.33	6.03 ^c ±0.62	5.52 ^b ±0.18	15.70 ^a ±0.05
FARO 57	90.65 ^b ±0.95	8.05 ^b ±0.38	5.19 ^c ±0.13	14.30 ^b ±0.17
NERICA 7	87.11 ^c ±0.74	10.80 ^a ±0.25	6.43 ^a ±0.14	13.10 ^c ±0.16
LSD	0.34	0.05	0.28	0.24

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each parameter is not significantly different (P>0.05)

Table 4.6: Sugar profile of barley and rice malt wort

SAMPLES	Maltotriose (%)	Glucose (%)	Maltose (%)	Maltotetraose (%)	Sucrose (%)	Raffinose (%)
BARLEYFARO	18.40 ^a ±0.07	10.75 ^b ±0.01	29.42 ^a ±0.01	1.05 ^a ±0.02	3.01 ^a ±0.02	0.11 ^a ±0.00
44	12.26 ^d ±0.28	11.23 ^a ±0.16	14.63 ^c ±0.11	0.63 ^b ±0.04	2.44 ^b ±0.10	0.07 ^a ±0.00
FARO57	13.25 ^c ±0.14	11.63 ^a ±0.71	15.34 ^b ±0.08	0.53 ^c ±0.03	2.32 ^c ±0.03	0.05 ^b ±0.00
NERICA 7	16.40 ^b ±0.07	10.84 ^b ±0.06	15.03 ^b ±0.04	0.44 ^d ±0.06	2.83 ^a ±0.08	0.06 ^{ab} ±0.00
LSD	0.03	0.02	0.03	0.03	0.03	0.01

Values are the means of duplicate determinations (N=2)

a,b,...means with the same superscript along a column for each parameter is not significantly different (P>0.05)

Table4.7: Measured values of analysed rice malt wort samples

Samples	pH	TSN (g/L)	Brix (g/100g)	KI (%)	FAN (mg/L)	DE (g/100g)	OE (g/100g)
BARLEY	5.60 ^a ± 0.42	5.70 ^a ±0.02	10.93 ^c ±0.30	45.15 ^a ±0.21	123. 64 ^a ±0.08	41.02 ^a ±0.61	11.21 ^a ±0.03
FARO 44	5.40 ^a ± 0.38	5.40 ^a ±0.00	14.65 ^b ±0.07	34.39 ^c ±0.13	108.56 ^d ±0.08	39.00 ^a ±1.42	11.15 ^a ±0.05
FARO 57	5.30 ^a ±0.17	5.60 ^a ±0.00	16.36 ^a ±0.42	39.16 ^b ±0.09	112.23 ^c ±0.28	40.00 ^a ±0.82	12.66 ^a ±0.04
NERICA 7	5.30 ^a ±0.10	5.80 ^a ±0.00	13.88 ^c ±0.11	44.27 ^a ±0.28	117. 34 ^b ±0.06	37.00 ^a ±0.01	9.68 ^a ±0.07
LSD	NS	NS	0.58	3.20	4.32	NS	NS

Values are the means of duplicate determinations (N=2)

a,b,....means with the same superscript along a column for each parameter is not significantly different (P>0.05)

TSN = Total soluble nitrogen; KI = Kolbach index; FAN = Free amino nitrogen;

DE = Dextrose equivalent; and OE= Original extract

Table 4.8: Measured values analysed rice malt beer samples

Samples	pH	Colour (°EBC)	Apparent Extract (g/100g)	Alcohol content (% ABV)	Turbidity (NTU)
BARLEY	4.20 ^a ±0.51	3.80 ^a ±0.13	5.33 ^a ±0.44	3.65 ^b ±0.03	3.20 ^a ±0.23
FARO 44	3.80 ^a ±0.44	3.70 ^a ±0.16	4.59 ^b ±0.74	3.54 ^b ±0.06	5.30 ^a ±0.01
FARO 57	3.90 ^a ±0.59	3.73 ^a ±0.71	4.57 ^b ±1.07	4.13 ^a ±0.18	4.80 ^a ±0.17
NERICA 7	3.80 ^a ±0.10	3.20 ^a ±0.42	4.93 ^b ±0.54	2.82 ^c ±0.50	4.30 ^a ±0.21
LSD	NS	NS	NS	0.39	NS

Values are the means of duplicate determinations (N=2)

a,b....means with the same superscript along a column for each parameter is not significantly different (P>0.05)

ABV = Alcohol byvolume

Table 4.9: Sensory attributes of rice malt beer of different varieties.

SAMPLE	Colour	Taste	Aroma	Mouthfeel	Appearance	Overall acceptability
BARLEY	8.71 ^a ±0.55	8.19 ^a ±0.13	7.99 ^a ±0.51	8.51 ^a ±0.25	8.61 ^a ±0.45	8.40 ^a ±0.38
FARO 44	6.91 ^b ±0.33	7.87 ^b ±0.32	7.81 ^a ±0.33	6.96 ^b ±0.32	6.52 ^b ±0.32	7.21 ^b ±0.30
FARO 57	6.66 ^b ±0.13	7.69 ^b ±0.21	7.54 ^a ±0.16	6.57 ^b ±0.41	6.24 ^b ±0.23	6.94 ^b ±0.08
NERICA 7	6.87 ^b ±0.25	7.82 ^b ±0.10	7.80 ^a ±0.51	6.76 ^b ±0.22	6.41 ^b ±0.23	7.13 ^b ±0.24
LSD	0.85	NS	NS	1.22	0.93	0.56

Values are the means of duplicate determinations (N=2)

a,b....means with the same supers

script along a column for each parameter is not significantly different (P>0.05)

4.2 DISCUSSION

4.2.1 Changes in Grain Quality of Rice Grain

4.2.2. Thousand Corn Weight

Rice grain varied significantly ($p < 0.05$) in TCW (25.81 – 27.01 g) across rice varieties (Table 4.1). Specifically, TCW, GE, and GC peaked at NERICA 7 with 27.01 g, 95.00 %, and 95.50 %, respectively. The range/variations of TCW, GE, and GC in this current study competes well with those reported by Osuji *et al.*, (2019), and could have arisen due to differences in soil composition, weather condition, moisture content or grain production/harvest period. Essentially, the TCW has been understood to help in identifying grain/seed density, size, and variety (Tokpah, 2010; Osuji *et al.*, 2019). Higher TCW or large grain size could also serve as an indicator of high starch content (Ayernor and Ocloo, 2007). On this premise, therefore, the NERICA 7 might have a higher starch content compared to other rice varieties in this current study.

4.2.3. Germinative Energy (GE) and Germinative Capacity (GC)

The rice grain's GE (86.50 – 95.00 %) and GC (92.50 – 95.50 %) differed significantly ($p < 0.05$) across rice varieties (Table 4.1). The germinative (GE and GC) outcomes of the rice grains of this current study seem favourable, and promises a good viability during malting (Adebowale *et al.*, 2010; Osuji *et al.*, 2019). Changes in GE and GC across the studied rice varieties might have

occurred with water absorption rates, TCW (kernel size), rice harvest period as well as starch content. Besides, the prompt grain germination of above 90 % of the grain would be considered a good quality malt attribute (Agbale *et al.*, 2007). In the current work, the rice varieties obtained acceptable grain germination of above 85 %, which makes it acceptable for malting purposes. Similar trends of over 85 % germinative properties were reported (Bam *et al.*, 2006; Hammond and Ayernor, 2001; Ameko *et al.*, 2013). Moreover, higher GE enhances the enzyme activities as well as seed vigour (Agbale, 2007). Additionally, both grain and malting qualities/characteristics are well-known to ascertain a cereal to become an acceptable substitute for barley. Therefore, the rice grain's GE (86.50 – 95.00 %) and GC (92.50 – 95.50 %) of current study shows a high promise as barley substitute in brewing.

4.2.4. Degree of Steeping (DoS)

The DoS (49.96 – 55.00 %) obtained significant differences ($p < 0.05$) across rice varieties (Table 4.1). DoS is the amount of water absorbed by the grain during steeping and it is an integral step in malting process because development of enzyme and formation, as well as metabolic transformation is influenced by it. In the current work, the DoS was highest (55.00 %) at FARO 44, and least at NERICA 7 (49.96 %). Results showed that rice varieties with higher DoS obtained higher MY and lower ML, probably due to decreased metabolic processes in the rice varieties of the current study. Besides small kernel staking

up more water compared to larger kernels, it is also believed that the grains from inland regions would swell and germinate faster compared to grains from maritime regions (Kunze, 2004).

4.2.5. Changes in Cold Water Extract (CWE), Hot Water Extract (HWE), Diastatic Power (DP), Malting Yield (MY), Malting Loss (ML), Moisture Content (MC) and Total Nitrogen (TN) of Rice Malt.

4.2.5.1 CWE, HWE and DP

Malt varied significantly ($p < 0.05$) in CWE (10.95 – 23.16 g/100g), HWE (51.06 – 206.48 L°/kg), and DP (22 – 150 °Lintner/kg) across rice varieties (Tables 4.2, 4.3 and 4.4). Specifically, CWE/HWE/DP peaked at FARO 44 (22.88 g/100g; 136.56 L°/kg; 150 °Lintner/kg), FARO 57 (22.13 g/100g; 170.18 L°/kg; 148 °Lintner/kg), and NERICA 7 (23.16 g/100g; 206.48 L°/kg; 143 °Lintner/kg), respective to $S_{18}G_3K_{55}^{\circ}$ (18 h steeping, 3 days germination, 55°C kilning), $S_{30}G_2K_{50}^{\circ}$ (30 h steeping, 2 days germination, 50°C kilning) and $S_{24}G_3K_{55}^{\circ}$ (24 h steeping, 3 days germination, 55°C kilning) combinations, and trended: FARO 44 ($S_{18}G_3K_{55}^{\circ}$) > FARO 57 ($S_{30}G_2K_{50}^{\circ}$) > NERICA 7 ($S_{24}G_3K_{55}^{\circ}$). Outside this (specific steeping duration, germination periods and kilning temperatures), CWE/HWE/DP produced no peaks (Tables 4.2, 4.3 and 4.4). Rice malt CWE (10.95 – 23.16 g/100g) range herein agrees with those reported by Kasetsart (2007). Based on CWE (15 - 22 g/100g) range data (Briggs 1998; Briggs *et al.*, 2004), rice malt herein represented a ‘good modification’. By hydrating the grist during grain modification, cold mashing

(20°C) solubilizes enzymatically-degraded compounds (Dahiya *et al.*, 2017). According to European Brewing Convention (EBC) and American Society of Brewing Chemists (ASBC), HWE range of 51.06 – 206.48 L°/kg parallel to ~13 – 54 g/100g soluble extract (SE) suggested HWE herein to be greater than two fold of CWE. Moreover, DP of rice malts (23 – 150 °Lintner/kg) corroborate with barley (50 – 150 °Lintner/kg) (BYO (2019a), (35 – 40 °Lintner/kg) (O'Rourke, 2002a) and sorghum (20 - 23°Lintner/kg) malts (Byrne *et al.*, 1993). Malt DP range 35 – 40 °Lintner/kg can convert its own starches (BYO, 2019b) probably with a longer conversion time. Malt enzymes that degrade starch and obtain high extract yield depict good malt characteristics (Sabramanian *et al.*, 1995; Muoria and Bechtel, 1998).

CWE, HWE and DP of rice malt increased with steeping duration at FARO 44 (S₁₈G₃K₅₅°), FARO 57 (S₃₀G₂K₅₀°) and NERICA 7 (S₂₄G₃K₅₅°) (Tables 4.2, 4.3 and 4.4). The positive correlation results [FARO 44 (R² = 0.769), FARO 57 (R² = 0.700) and NERICA 7 (R² = 0.588)] show DP's significant role in HWE of rice malt, corroborating with (correlation) data of millet malt (Eneje, *et al.*, 2012). Besides small-sized kernel of cereals modifying at a faster rate over large ones (Agu, 2009), the grain physiological activities progressing during malting (Kunze, 2004; Ogbonna, 2002; Osujiet *et al.*, 2019) can influence CWE, HWE and DP of malt. Starch-degrading enzymes like alpha-amylase, beta-amylase, limit dextrinase and alpha-glucosidase (Buchholzet *et al.*, 2005; Evans *et al.*, 2010), lipases, proteases and other enzymes could also influence DP of malt (Briggs,

1998). Low DP of rice malts corroborate with lower protein content of grain (Agu and Palmer, 1998). Mashing schedule, high gelatinization temperature, and kilning/malting conditions might be contributing to differences in CWE, HWE and DP of rice malt in this study.

4.2.5.2. Malt Yield, Malting Loss, Moisture Content and Total Nitrogen

To a maltster, MY is an important attribute because it gives an indication of the amount of recoverable soluble extracts from the malted grain (Osuji *et al.*, 2019). Additionally, the malt quality analyses guide the maltster/brewer on both variety selection and effectiveness of malting process for optimized output, to achieve sustainable malt brewing process (Briggs, 1998). On the other hand, ML is the measure of metabolic activity associated with grain germination, which increases with germination period. In the current work, the MY (87.11 – 92.65 %) and ML (6.03 – 10.80 %) of rice malt changed significantly ($p < 0.05$) across varieties (Table 4.5). Peak (10.80 %) ML and least (87.11 %) MY can be seen at NERICA 7 while Peak MY (92.65 %) and least (6.03 %) ML can be seen at FARO 44. Probably, these variations in ML and MY might have been influenced by the malting process (Ofoedu, 2019; Osuji *et al.*, 2019). Besides moisture loss during kilning as well as physiological activities associated with germination, the changes in TCW, GE and GC, would perhaps influence both MY and ML. We opine that this might be so, considering the peak TCW, GE, and GC values obtained at NERICA 7, as well as the least MY obtained at FARO 44.

MC and TN of rice malts specific to FARO 44 (S₁₈G₃K₅₅^o), FARO 57 (S₃₀G₂K₅₀^o) and NERICA 7 (S₂₄G₃K₅₅^o) were determined. MC (5.19 – 6.43g/100g) and TN (13.10 – 15.70g/L) varied significantly (p<0.05), with the following trends for MC (NERICA 7 =6.43g/100g > FARO 44= 5.52g/100g > FARO 57= 5.19g/100g) and TN (FARO 44 = 15.70g/L > FARO 57 = 14.30g/L > NERICA 7 =13.10g/L) (Table 4.5). MC of rice malt (5.19 – 6.43g/100g) was above those of Munich (3.0 – 4.8g/100g) as well as two-row (2.0 – 4.3g/100g) barleys (Noonan, 2003). Increase in MC of malts can decrease the extract potential, which might lower original gravity of wort (BYO, 2019a). Malt closer to 1.5g/100g MC is of less risk to the mould growth (Noonan, 2003). Moisture level of grain is reduced with high drying temperature(s) or prolonged drying time(s) (Osuji *et al.*, 2019). Whereas low MC can prolong food shelf life (Alozie *et al.*, 2009), high MC can enhance its microbial spoilage (Ijarotimi, 2012). TN of rice malt (13.10 – 15.70g/L) fell within those of ale/lager (14.00 – 18.00g/L) (O'Rourke, 2002a), and sorghum (14.70 – 17.40g/L) types (Agu and Palmer, 1998). Rice variety as well as malting conditions may contribute to the MC and TN differences in rice malt of this study. As nitrogen via amino acids is required for yeast growth, hydrophobic nitrogen (from malt) provider foam and head retention in beer (O'Rourke, 2002a).

4.2.6. Changes in Sugar profile of Rice Malt Wort

To the brewer, the importance of sugar wort parameter/composition underpins the influential success of fermentation. Additionally, the sugar profile of rice malt wort is the outcome of the enzymatic activities during mashing. In the current work, the rice malt wort yielded a combination of sugars, such as maltose (14.63 – 15.34 %), maltotriose (12.26 – 16.40 %), glucose (10.84 – 11.63 %), sucrose (2.32 – 2.83 %), raffinose (0.05 – 0.07 %) and maltotetraose (0.44 – 0.63 %), all of which varied significantly ($p < 0.05$) across varieties (Table 4.6). Clearly, it was not difficult to differentiate the sugars herein based on amounts obtained across the studied rice varieties. Specifically, whereas the maltose, glucose and maltotriose clearly obtained higher amounts, the sucrose, maltotetraose and raffinose obtained lower amounts. Both maltose and maltotriose, well-known as predominant sugars found in wort are, seemed to be noticeably lower than the values obtained by Ofoedu *et al.*, (2019).

Malting conditions, mashing program as well as the nature/type of (exogenous) enzymes used could have some impact on the variations in the rice malt wort sugar profile. Additionally, the lower sugar concentrations in the rice malt wort might be attributable to limit dextrins produced in higher amounts, and maybe, tannins binding with malt's amylase enzyme (Okolo *et al.*, 2010). Sucrose, among the major soluble sugars and natural component of matured kernel, is neither produced during malting nor hydrolysis/mashing, but however, could be depleted naturally during germination in sustaining (rice) malt metabolism. This

might explain the significantly low sucrose concentration in the rice malt wort. The presence of maltotetraose and raffinose in the wort could be an indication of oligosaccharides resulting from limit dextrans formation due to the different amylolytic enzymes working in the rice malts (Marconi *et al.*, 2017).

4.2.7 Changes in pH, Total Soluble Nitrogen(TSN), Brix, Kolbach Index(KI), Free Amino Nitrogen(FAN), Dextrose Equivalent(DE) and Original Extract(OE) of Rice Malt Wort.

4.2.7.1. pH and Total Soluble Nitrogen.

Resembling ($p>0.05$) across rice varieties (FARO 44 = 5.40 > FARO57/NERICA 7 = 5.30), pH of wort (Table 4.5) compares well with another previously published (rice wort) data (4.98 – 6.08) (Kasetsart, 2007) but slightly below those of barley ale/lager (5.6 - 5.9) (Palmer, 2006). During mashing and wort boiling, heat treatment can dissociate the calcium ion (Ca^{2+}) bound with both phosphates $\text{S}(\text{K}_2\text{PO}_4)$ and polypeptides, forming insoluble compounds, releasing hydrogen ion (H^+), and decreasing wort pH (Palmer, 2006). Increased wort acidity enhances both protein coagulation and yeast growth, and inhibits microbial contamination (O'Rourke, 2002b).

TSN of rice malt wort resembled ($p>0.05$) across rice varieties (NERICA 7= 5.80g/L > FARO 57=5.60g/L> FARO 44=5.40g/L (Table 4.7), with the range (5.40 – 5.80g/L) corroborating favourably with those of sorghum (5.00 – 7.00g/L) (Agu & Plamer, 1998) and typical lager barleys (5.70 – 6.60g/L) (O'Rourke, 2002a). Aided by denaturation and precipitation of solubilized

proteins, high gelatinization temperature of rice starch reduces TSN level in wort. Steeping could enhance the loss of some soluble nitrogenous compounds, like amino acids (Briggs, 1998). Amino acid dissolution could increase in TSN (during germination) (Banusha and Vasantharuba, 2013), which would cease if acrospires reach from 3/4 to 7/8 of grain length (Briggs et al., 2004). As the need of wort TSN increases, it becomes undesirable if protein degradation raises the TSN levels higher than required, to reduce foam formation, abnormal fermentation (Sadosky, 2007), and haze formation (Briggs *et al.*, 2004).

4.2.7.2 Brix and Kolbach Index

Brix of rice malt wort differed significantly ($p < 0.05$) across rice varieties (FARO 57 =16.36g/100g > FARO 44 =14.65g/100g > NERICA 7 =13.88g/100g) (Table 4.7). Peak brix at FARO 57 suggested increased malting accessibility to substrate (starch) with enhanced enzymatic hydrolysis. Grain kernel size differences in rice varieties might affect the endosperm starch composition when malted/mashed, which may well vary the brix values. Varying malting conditions influencing how the grain responds to its modification, could differ during germination. Brix value could, therefore, be affected by amount/type of sugars in wort, which serves as a nutrient for yeast (Pedley, 1996).

KI of rice malt wort differed significantly ($p < 0.05$) across rice varieties (NERICA 7 =44.27% > FARO 57 =39.16% > FARO 44 =34.39%) (Table 4.7) with (34.39 – 44.27%) resembling those of typical lager malt (34 - 44%)

(O'Rourke, 2002a). Malts can be classified, based on degree of modification (BYO, 2019b), namely: a) under modified (KI values between 30 – 35%), b) well modified (KI values above 35%), c) over modified (KI values above 45%) malts. Specifically, wort KI of FARO 44 (34.39%) falls within 'under modified', whereas FARO 57 (39.16%) and NERICA 7 (44.27%) falls within 'well modified' malts. Further, Bamforth (2003) reported 'well modified' malt with KI range of 38 – 42%. Besides malting enabling KI to increase with germination, the small-size of FARO 44 kernel may corroborate with lower (KI) value. Thinner kernel/grain size taking up water faster (Kunze, 2004; Osuji et al., 2019) might sustain a higher TN relative to the larger ones (Briggs, 1998). The reduced HWE, CWE and KI values might help in defining those of FARO 44 as 'under modified malt'. However, the higher TSN (NERICA 7 = 5.80g/L) and KI values (NERICA 7 = 44.27%) would suggest a positive association of grain size with HWE, CWE and TSN of the current study.

4.2.7.3. Free Amino Nitrogen, Dextrose Equivalent and Original Extract

FAN of rice malt wort differed significantly ($p < 0.05$) across rice varieties (NERICA 7 = 117.34mg/L > FARO 57 = 112.23mg/L > FARO 44 = 108.56mg/L) (Table 4.7) and compared well with those of sorghum (94 – 216 mg/L) (Agu & Palmer, 1998), maize (100 – 169mg/L) and rice (95 – 138mg/L) (Taylore *et al.*, 2013) malts. Increased amino acids/protein modification might favour the peak FAN in NERICA 7 with proteolytic enzyme activity. As principal nitrogen source in wort, FAN depicted hydrolysed (soluble) proteins during mashing

(Agu and Palmer, 1998; Russell, 2006), summed up by amino acids, ammonium ions and small peptides (dipeptides and tripeptides) (Stewart *et al.*, 2013; Lekkas *et al.*, 2005; Pugh *et al.*, 1997). Typical lager malt with FAN between 100 and 140mg/L can enhance efficient yeast cell growth and fermentation performance (Lekkas *et al.*, 2005) to achieve a trouble-free fermentation (Briggs *et al.*, 2004). Besides, FAN can also help to predict yeast's healthy growth, viability/vitality, and fermentation efficiency (Hill & Stewart, 2019). Though FAN strongly depends on malting conditions (Briggs, 1998), some FAN components (alongside reducing sugars) during mashing might provide minor flavour precursors that undergo maillard reaction (Hill and Stewart, 2019; Hughes, 2009). Despite FAN influencing other fermentation factors (like cell biomass, growth, pH, viability, and attenuation rate) (Shimizu *et al.*, 2002), too high FAN is undesirable given the resultant excessive yeast growth, which could affect beer stability (BYO, 2019b). Malts with higher FAN levels require adjuncts, which can act as nitrogen diluent, but would contribute little-to-no TSN to the wort (Briggs *et al.*, 2004).

Resembling ($p > 0.05$) across rice varieties (FARO 57 = 40g/100g > FARO 44 = 39g/100g > NERICA 7 = 37g/100g), the peak DE of rice wort (Table 4.7) suggested increased hydrolysis. The slight DE variations in wort might reflect the differences in amylose-amylopectin ratio of rice starch. This is because the amylose can be more completely hydrolyzed than amylopectin, the latter limited by beta-limit dextrin due to branched chains (Osuji and Anih, 2011). Varietal

differences, varying malting conditions as well as amount/type of enzymes developed in the rice grain particularly during malting might also govern both degree of hydrolysis and hydrolysates types obtained. The maltose and maltotriose remain the most abundant sugars present in malt/wort (Goldhammer, 2008; Palmer, 2009), which could also influence the DE of wort (Ofoedu *et al.*, 2020).

Resembling ($p > 0.05$) across rice varieties (FARO 57 = 12.66g/100g > FARO 44 = 11.15 g/100g > NERICA 7 = 9.68g/100g), OE of rice malt wort (Table 4.7), neared those of millet (~10 g/100g), sorghum (10.42g/100g) and barley (11.0 g/100g) malts (Reginald, 1995), and compared well with other reported ranges (7.5 - 9, 8 - 9.5, 11-14, and 12.5 - 16 g/100g) of different barleys used for ale beers (Papazian, 2006). Principally, original gravity (density) of wort is four times the original extract by Plato scale. Well-known, water density is 1.0000 at standard temperature and pressure (STP); if respective wort density of FARO 44, FARO 57 and NERICA 7 were 1.04448 (11.15 g/100g), 1.05064 (12.66 g/100g) and 1.03872 (9.68 g/100g), the corresponding wort will be 44.48°, 50.64° and 38.72° of excess gravity. Thus, wort densities consider the solution factors/mixtures of dissolved carbohydrate materials, soluble proteins and minerals typically emerging from malted cereal materials. Besides grain mashing to considerably influence OE of wort, most grain modified products (that is, cell wall degradation and enzymatic breakdown) in endosperm's

protein-starch matrix (Agu and Palmer, 2001) would be released (into the wort) as soluble extracts.

4.2.8 Changes in pH, Colour, Apparent Extract(AE), Alcohol by Volume(%ABV), Turbidity, and Sensory Attributes of Rice Malt Beer

4.2.8.1 pH, Colour and AE

Resembling ($p > 0.05$) across rice varieties (FARO 57 = 3.90 > FARO 44 & NERICA 7 = 3.80), pH of beer (Table 4.8) appeared lower than those of barley (4.1 – 4.5) (Bamforth, 2001) as well as sorghum (3.90 – 4.10) beers (Iwouno *et al.*, 2019). Low pH in rice malt beer might be due to organic (weak) acids excreted by yeast with excess CO₂ (which provides relative amounts of carbonic acid) during fermentation. Low beer pH also depicts its sharpness of taste. When pH is below 4, taste further sharpens with increased foam stability and head retention (Bamforth, 2001). By decreasing buffering capacity, lower pH increases yeast growth, removes colloidal particles of proteins-polyphenol complexes (and other insoluble materials) and inhibits microbial growth (in beer/wort) (Leiper and Miedl, 2006). In addition, pH in beer, determined by organic acids, e.g., acetic, lactic, pyruvic and citric acid, can influence its flavour.

Beer colour depicts its appearance and its critical to (product) acceptance, being the first quality attribute consumers perceive (Leonet *al.*,2006; Osujiet *al.*, 2020). Beer colour resembled ($p>0.05$) across rice varieties (FARO 57= 3.73 °EBC > FARO 44= 3.70 °EBC > NERICA 7= 3.20 °EBC) (Table 4.8). Although fairly above those of another rice malt (1.70 – 2.60 °EBC) (Mayer *et al.*, 2017), rice malt beer herein compared well with those of typical barley (2.00 – 4.00 °EBC) (O'Rourke, 2002a), but not so for sorghum (6.0 – 6.6 °EBC), barley double crown (~7.5 °EBC) and barley rex (~14.0 °EBC) malt lager beers (Olatunji *et al.*, 1993). As °EBC increases, beer colour darkens. When assessed by Saveur Bierre colour chart (Anon., 2020), rice malt beer colour range herein depicted type pale yellow lager. Beer colour variations could be as a result of either decolourization of the (beer colour) substance as pH dropped (Kunze, 2004), changes/differences in malt colour, or inconsistencies in the colour formation of wort during boiling process (Briggs *et al.*, 2004). Phenols (tannins) are natural organic compounds in malts/hops, which change beer colour from pale yellow to dark brown via Maillard reaction/caramelization (Whistler and Bemiller, 2008; Panthare*et al.*,2013). In addition, maillard reaction and caramelization occurring independently/simultaneously would influence colour formation/intensity (Kunze, 2004). Other factors like pH level, yeast strain, hop usage, maturation duration and specialty ingredients can influence beer colour.

AE resembled across rice varieties ($p>0.05$) (NERICA 7=4.93 g/100g > FARO 44 = 4.59 g/100g > FARO 57 = 4.57 g/100g) (Table 4.8). Noticeably, there appears some reduction in gravity of wort from 9.68 – 12.66 g/100g (Table 4.4) to 4.57 – 4.93 g/100g (Table 4.5) in the final rice malt beer. Dissolved solids (sugars, amino acids, minerals, among others) in wort utilized by yeast during fermentation might reduce the final beer gravity. As yeast utilizes sugars (and other compounds) to produce alcohol, the gravity of wort may well decrease (Boulton, 1991; Briggs *et al.*, 2004). Moreover, fermentability of wort depicts the proportion of dissolved solids (extract) that can be fermented. In other words, 59 % (FARO 44), 64 % (FARO 57) and 49 % (NERICA 7) of fermentable materials in these wort utilized by yeast produced AE of 4.59 g/100g, 4.57 g/100g and 4.93 g/100g, respectively.

4.2.8.2. Alcohol Content and Turbidity

Alcohol content of beer, although differing significantly ($p<0.05$) across rice varieties (FARO 57 =4.13%ABV > FARO 44 =3.54 %ABV> NERICA 7 =2.82 %ABV) (Table 4.8), fell within a generally anticipated range (4 – 6 %ABV) (Polanet *al.*, 2015), somewhat above 2.55, 3.09 and 3.65 %ABV of millet, sorghum and barley beers, respectively (Reginald, 1995). FARO 57 with peak fermentability of 64% corresponded to 4.13 %ABV, and NERICA 7 with least fermentability of 49% corresponded to 2.82 %ABV. This suggested alcohol concentration in beer not solely dependent on the original extract/gravity of wort, but more likely on the availability of fermentable extracts, readily utilized

by the yeast. Whilst the fermentable extracts especially sugars in wort remain the beer quality index (Jordao *et al.*, 2015), its concentration (and subsequent utilization) in the wort can help to determine the improved fermentation efficiencies (Zhao *et al.*, 2008).

Resembling ($p > 0.05$) across rice varieties (FARO 44 = 5.30 NTU > FARO 57 = 4.80 NTU > NERICA 7 = 4.30 NTU), beer turbidity (Table 4.8) were above those of sorghum (1.6 – 2.0 NTU) and barley malt (3.2 NTU) lager beer (Olatunji *et al.*, 1993), but below those of sorghum (red) (South Africa) (~12.8 NTU), sorghum (white) (Australia) (~28 NTU), sorghum (white) (Nigeria) (~33.2 NTU) malt beers (Aisen and Muts, 1987), and sorghum beer clarified with different filter aids (8.28 – 26.56 NTU) (Iwouno *et al.*, 2019). Considering 1.00 EBC equals 4.00 NTU, the beer turbidity can be graded based on degree of haziness, which includes; brilliant: 0 – 0.50 EBC (0 – 2.00 NTU); almost brilliant: 0.50 – 1.00 EBC (2.00 – 4.00 NTU); very slightly hazy: 1.00 – 2.00 EBC (4.00 – 8.00 NTU); slightly hazy: 2.00 – 4.00 EBC (8.00 – 16.00 NTU); hazy: 4.00 – 8.00 EBC (16.00 – 32.00 NTU) and very hazy: > 8.00 EBC (> 32.00 NTU) (Callemien and Collin, 2009). Herein, the rice malt beer (4.30 – 5.30 NTU) would be considered as ‘very slightly hazy’. Some proteins not removed during wort boiling, surviving fermentation, and finding its way into the beer might equally cause the haze (Briggs *et al.*, 2004). Besides the origin of haze formation as either biological (e.g., bacteria, cell debris, yeast) or non-

biological (inorganic, carbohydrate-based and protein-polyphenol complexes) (Siebert *et al.*, 1996; Stewart, 2004; Briggs *et al.*, 2004), beer haziness might be due to ineffective filtration, non-flocculent yeast, and or poorly modified malt/filter aids (Steiner *et al.*, 2010). Coloured compounds such as melanoidins (Iwouno *et al.*, 2019), cereal/malt-type, and differences in chemical composition/processing methods can influence beer turbidity. In addition, centrifugation and microfiltration used during commercial production can also increase beer clarity (Kuiper *et al.*, 2002; Shotripuk *et al.*, 2005).

4.2.8.3 Sensory Attributes

The sensory attributes (colour, aroma, taste, mouthfeel, appearance, and overall acceptability) of rice malt beers compared with commercial lager beer is shown in Table 4.9. The colour (6.66 – 8.71), mouthfeel (6.57 – 8.51), appearance (6.24 – 8.61) and overall acceptability (6.94 – 8.40) of beer samples differed significantly ($p < 0.05$), but not for taste (7.69 – 8.19) and aroma (7.54 – 7.99) ($p > 0.05$). Based on the hedonic scale, the panellists viewed the colour of the rice malt beers (6.66 – 6.91) as pale yellow colour and compared to the commercial lager beer (8.71). The panellists considered the rice malt beers as slightly liked compared to the commercial lager beer that was liked very much. The colour variations in the beer samples may be due to differences in kilning temperatures and chemical compositions (sugars and amino acids) that facilitates formation of melanoidin in beer (Osuji *et al.*, 2020; Iwouno *et al.*, 2019). The

panellists obtained mouthfeel of rice malt beers (6.57 – 6.96) as slightly liked/relatively flat compared to the commercial lager beer (8.51) which was liked very much. The variations in mouthfeel of beer samples may be due to varying concentrations of residual sugars, higher alcohols as well as organic acids in the beer (He *et al.*, 2014; Iwouno *et al.*, 2019).

Appearance, which also include but not limited to colour and absence of haze greatly affect beer perception. Like mouthfeel and colour of beer samples, the appearance (6.24 – 6.52) of rice beer samples was slightly liked probably because the rice malt beers appeared very slightly hazy compared to the commercial lager beer (8.61) which appeared almost brilliant in clarity. The variations in appearance could be due to differences in brewing technology adopted. Notably, the taste and aroma across the beer samples resembled ($p > 0.05$), although the sensory scores indicated the taste and aroma of commercial lager beer as liked very much, whereas that of rice malt beer samples were liked moderately. Specifically, aroma and taste of beer is characterized by volatile compound profile (Marconi *et al.*, 2017) influenced principally by yeast metabolism. The differences in taste and aroma of beer samples may occur with fermentation by-products, such as aroma-active esters, higher alcohols, and aldehydes (He *et al.*, 2014; Ferreira and Guido, 2018). The overall acceptance herein suggests sensory properties of beer might be affecting consumer liking, considering the commercial lager beer was liked very much by the panelists. Besides FARO 44 and NERICA 7 beer being liked moderately, the

FARO 57 beer was slightly liked. Overall, the rice malt beer resembled in sensory profiling to the commercial lager beer in aroma and taste, but more flat in mouthfeel.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

The rice varieties exhibited desirable grain quality characteristics and showed acceptable aptitude to be malted due to their germinative property of greater than 85 %. Malting conditions significantly influenced the Cold Water Extract, Hot Water Extract, Diastatic Power, Moisture Content and Total Nitrogen of rice malt. Through peak FARO 44 (S₁₈G₃K₅₅°), FARO 57 (S₃₀G₂K₅₀°) and NERICA 7 (S₂₄G₃K₅₅°) malt outputs, the resulting wort produced promising pH, Total Soluble Nitrogen, Brix, Kolbach Index, Free Amino Nitrogen, Dextrose Equivalent, Original Extract ranges and sugar profile similar to that of barley malt wort. Across varieties, the pH, Total Soluble Nitrogen, Brix, Kolbach Index, Free Amino Nitrogen, Dextrose Equivalent and Original Extract in rice malt wort, and pH, colour, Apparent Extract and turbidity in rice beer resembled (p>0.05), but not so in ABV (p<0.05). In addition, the rice malt beer, very slightly hazy, represented a pale-yellow light lager. To obtain wort that makes

an alcohol clear beer requires the addition of exogenous enzymes, particularly in the mashing of rice malts. Moreover, malting would improve hydrolysis, modify the starchy (rice) endosperm, which could allow adequate production of Free Amino Nitrogen, Total Soluble Nitrogen and other fermentable extracts in wort. Besides the sensory profiling differing in appearance, the characteristic pale yellow rice malt beer resembled the commercial lager beer in aroma and taste, but more flat in mouthfeel. However, the overall acceptance suggests that rice malt beer from FARO 44 was preferred more amongst other rice malt beers, after the commercial lager beer. Malting conditions of current study appears very promising for commercial lager beer production since not all the variety is suitable for brewing, and therefore careful variety selection is for optimized output is important.

5.2 Recommendation.

Further research is warranted on other locally available rice varieties as well as underutilized cereals for malting/brewing, which would target higher extract yield as well as a clearer beer. However, there is a chance that not all the local rice varieties (in Nigeria and other rice thriving West African countries) would be suitable for brewing. Therefore, a careful and thorough rice variety selection process would be needful if an optimized malt beer output is to be actualized.

Another research direction of future studies could be to determine the foam formation, retention, bubble size, and distribution as well as microbiological analysis of rice malt beer subject to selected/varying malting conditions.

Additionally, future studies could quantitatively and qualitatively determine and characterize enzymes in rice malts of different varieties as influenced by varying malting conditions.

The sensory profile can further be improved by using high kilning temperature and different yeast strain, hop variety or proportion of ingredients.

The findings of this research work will find wide applications in the brewery industries especially for tropical beer brewing using indigenous rice varieties.

5.3 Contribution to Knowledge

Modifying malting conditions enhances development of adequate endogenous enzyme in the grist thereby significantly reducing the amount of exogenous enzyme used during mashing operation.

Malted rice from some locally produced rice varieties has been fully explored as a principal raw material for brewing.

These added values of rice could increase rice production quest to attain self-sufficiency in rice production

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APPENDIX

CWE, HWE and DP of FARO 44 VARIETY

Malting conditions			Malt analysis					
Steeping duration (hours)	Germination period (days)	Kilning temperature (°C)	CWE		HWE		DP	
			1 st	2 nd	1 st	2 nd	1 st	2 nd
18	2	50	21.68	21.64	121.90	121.22	78.72	77.68
		55	22.32	22.24	103.90	102.46	92.72	91.88
	3	50	21.91	21.77	103.70	102.66	62.70	62.30
		55	22.97	22.79	137.38	135.74	150.17	149.84
	4	50	20.51	20.39	106.64	105.80	80.26	79.74
		55	20.65	20.41	116.13	114.99	98.60	98.00
24	2	50	20.87	20.67	100.95	99.35	42.94	42.86
		55	21.05	20.85	85.56	84.38	63.33	63.07
	3	50	20.66	20.50	100.27	100.03	50.15	49.85
		55	20.80	20.72	91.35	90.73	52.40	52.00
	4	50	19.87	19.75	85.35	84.59	43.84	43.36
		55	20.55	20.43	106.38	105.98	57.53	56.67
30	2	50	18.53	18.47	51.29	50.83	37.61	37.40
		55	19.81	19.63	76.15	75.81	38.97	38.43
	3	50	19.71	19.51	76.44	75.52	35.48	35.12

	55	20.61	20.57	97.45	96.67	37.73	37.28
4	50	18.54	18.46	60.85	60.55	39.06	38.74
	55	20.52	20.40	94.65	93.77	40.27	39.93

CWE, HWE and DP of FARO 57VARIETY

Malting conditions			Malt analysis					
Steeping duration (hours)	Germination period (days)	Kilning temperature (°C)	CWE		HWE		DP	
			1 st	2 nd	1 st	2 nd	1 st	2 nd
18	2	50	19.45	19.41	103.34	102.76	63.62	62.78
		55	14.42	14.36	100.85	99.45	22.40	22.20
	3	50	14.03	13.95	105.70	104.64	50.16	49.84
		55	11.48	11.40	103.80	102.56	37.54	37.46
	4	50	16.56	16.46	161.07	159.79	37.62	37.38
		55	14.46	14.30	105.69	104.65	26.18	26.02
24	2	50	20.68	20.48	107.07	105.37	80.08	79.92
		55	15.24	15.10	103.90	102.46	38.78	38.62
	3	50	18.31	18.17	106.35	106.09	63.99	63.61
		55	13.23	13.17	106.58	105.86	52.40	52.00
	4	50	16.96	16.86	106.70	105.74	57.41	56.79
		55	17.23	17.13	103.68	103.28	39.00	38.42
30	2	50	22.17	22.09	170.96	169.40	148.41	147.59
		55	16.59	16.43	106.46	105.98	43.20	42.60
	3	50	19.07	18.89	167.91	165.91	135.98	134.62
		55	15.19	15.15	137.11	136.01	98.67	97.49

4	50	19.07	18.99	106.27	105.77	92.67	91.93
	55	16.56	16.46	103.56	102.60	42.78	42.42

CWE, HWE and DP of NERICA 7 VARIETY

Malting conditions			Malt analysis					
Steeping duration (hours)	Germination period (days)	Kilning temperature (°C)	CWE		HWE		DP	
			1 st	2 nd	1 st	2 nd	1 st	2 nd
18	2	50	10.96	10.94	136.94	136.18	71.07	70.13
		55	12.71	12.65	183.37	180.83	97.03	96.17
	3	50	11.77	11.69	164.53	162.89	48.15	47.85
		55	13.36	13.26	183.19	181.01	60.07	59.93
	4	50	12.72	12.64	121.90	120.92	66.91	66.49
		55	20.37	20.13	164.70	163.10	100.31	99.69
24	2	50	12.58	12.46	168.25	165.57	98.10	97.90
		55	15.01	14.87	149.04	146.96	120.24	119.76
	3	50	12.57	12.47	182.32	181.88	60.18	59.82
		55	23.21	23.11	207.18	205.78	143.56	142.44
	4	50	12.97	12.89	182.92	181.28	60.32	59.68
		55	21.19	21.07	133.95	133.45	101.26	99.74
30	2	50	13.34	13.30	140.24	138.96	102.29	101.71
		55	17.27	17.11	127.79	127.21	113.79	112.21
	3	50	13.57	13.43	128.27	126.74	92.77	91.85
		55	14.55	14.53	164.56	163.24	138.83	137.17

4	50	13.72	13.66	173.40	172.56	46.38	46.02
	55	16.39	16.29	134.33	133.07	75.32	74.69

Sugar Profile of rice malt wort

Sample	Maltotriose		Glucose		Maltose		Maltotetraose		Sucrose		Raffinose	
	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>
FARO 44	12.06	12.46	11.12	11.34	14.55	14.71	0.60	0.66	2.37	2.51	0.07	0.07
FARO 57	13.15	13.35	11.13	12.13	15.28	15.4	0.51	0.55	2.30	2.34	0.05	0.05
NERICA 7	16.35	16.45	10.8	10.88	15.00	15.06	0.40	0.48	2.77	2.89	0.06	0.06

Grain quality characteristics of different rice varieties

Sample	TCW		GE		GC		DoS	
	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>
FARO 44	25.80	25.82	86.46	86.54	92.40	92.60	54.80	55.02
FARO 57	26.00	26.22	93.45	93.55	93.00	93.00	50.90	51.00
NERICA 7	26.96	27.06	94.30	95.70	95.43	95.57	49.90	50.02

Measured values of analysed of rice malt samples

Sample	MY		ML		MC		TN	
	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>
FARO 44	92.88	92.42	6.47	5.59	5.65	5.39	15.74	15.66
FARO 57	91.32	89.98	8.32	7.78	5.28	5.10	14.42	14.18
NERICA 7	87.63	86.59	10.62	10.98	6.53	6.33	13.21	12.99

Measured values of analysed rice malt wort samples

Sample	pH		TSN		Brix		KI		FAN		DE		OE	
	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>	<i>1st</i>	<i>2nd</i>
FARO 44	5.67	5.13	5.40	5.40	14.60	14.70	34.30	34.48	108.50	108.62	38	40	11.11	11.19
FARO 57	5.42	5.18	5.60	5.60	16.06	16.66	39.10	39.22	112.20	112.60	42	38	12.63	12.69
NERICA 7	5.37	5.23	5.80	5.80	13.80	13.96	44.07	44.47	117.30	117.38	37	37	9.63	9.73

FARO 44 Sensory evaluation score sheet

Number of panelist	Colour	Taste	Aroma	Mouthfeel	Appearance	Overall acceptability
1	7.45	8.26	8.35	7.50	6.91	7.69
2	7.03	8.05	7.93	7.08	6.70	7.36
3	7.41	8.06	8.31	7.46	6.71	7.59
4	7.15	8.02	8.05	7.20	6.67	7.42
5	7.35	8.03	8.25	7.40	6.68	7.54
5	7.22	8.04	8.12	7.27	6.69	7.47
6	7.13	8.20	8.03	7.18	6.85	7.48
7	7.03	8.23	7.93	7.08	6.88	7.43
8	7.00	8.39	7.90	7.05	7.04	7.48
9	7.05	8.01	7.95	7.10	6.66	7.35
10	7.12	8.26	8.02	7.17	6.91	7.50
11	6.35	7.44	7.25	6.40	6.09	6.71
12	6.77	7.65	7.67	6.82	6.30	7.04
13	6.39	7.64	7.29	6.44	6.29	6.81
14	6.65	7.68	7.55	6.7	6.33	6.98
15	6.45	7.67	7.35	6.50	6.32	6.86
16	6.58	7.66	7.48	6.63	6.31	6.93
17	6.67	7.50	7.57	6.72	6.15	6.92
18	6.77	7.47	7.67	6.82	6.12	6.97
19	6.80	7.31	7.70	6.85	5.96	6.92
20	6.75	7.69	7.65	6.80	6.34	7.05

FARO 57 Sensory Evaluation Score sheet

Number of panelist	Colour	Taste	Aroma	Mouthfeel	Appearance	Overall acceptability
1	7.20	7.15	7.00	6.96	5.70	6.80
2	6.78	7.57	7.42	6.75	6.12	6.93
3	7.16	7.19	7.04	6.76	5.74	6.78
4	6.90	7.45	7.30	6.72	6.00	6.87
5	7.10	7.25	7.10	6.73	5.80	6.80
5	6.97	7.38	7.23	6.74	5.93	6.85
6	6.88	7.47	7.32	6.90	6.02	6.92
7	6.78	7.57	7.42	6.93	6.12	6.96
8	6.75	7.60	7.45	7.09	6.15	7.01
9	6.80	7.55	7.40	6.71	6.10	6.91
10	6.87	7.48	7.33	6.96	6.03	6.93
11	6.24	8.11	8.10	6.14	6.66	7.05
12	6.45	7.90	7.68	6.35	6.45	6.97
13	6.44	7.91	8.06	6.34	6.46	7.04
14	6.48	7.87	7.80	6.38	6.42	6.99
15	6.47	7.88	8.00	6.37	6.43	7.03
16	6.46	7.89	7.87	6.36	6.44	7.00
17	6.30	8.05	7.78	6.20	6.60	6.99
18	6.27	8.08	7.68	6.17	6.63	6.97
19	6.11	8.24	7.65	6.01	6.79	6.96
20	6.49	7.86	7.70	6.39	6.41	6.97

NERICA 7 Sensory Evaluation Score sheet

Number of panelist	Colour	Taste	Aroma	Mouthfeel	Appearance	Overall acceptability
1	7.13	8.08	7.99	7.03	6.59	7.36
2	6.98	7.93	8.00	6.88	6.60	7.28
3	7.36	8.31	8.01	7.26	6.61	7.51
4	7.10	8.05	7.97	7.00	6.57	7.34
5	7.09	8.04	7.98	6.99	6.58	7.34
5	7.17	8.12	7.99	7.07	6.59	7.39
6	7.08	8.03	8.15	6.98	6.75	7.40
7	6.98	7.93	8.18	6.88	6.78	7.35
8	6.95	7.90	7.94	6.85	6.54	7.24
9	7.00	7.95	7.96	6.90	6.56	7.27
10	7.07	8.02	7.99	6.97	6.59	7.33
11	6.66	7.61	7.52	6.47	6.21	6.97
12	6.65	7.60	7.67	6.62	6.20	7.03
13	6.64	7.59	7.29	6.24	6.19	6.87
14	6.68	7.63	7.55	6.50	6.23	6.99
15	6.67	7.62	7.56	6.51	6.22	6.99
16	6.66	7.61	7.48	6.43	6.21	6.95
17	6.50	7.45	7.57	6.52	6.05	6.96
18	6.47	7.42	7.67	6.62	6.02	6.99
19	6.71	7.66	7.70	6.65	6.26	7.05
20	6.69	7.64	7.65	6.60	6.24	7.03

Commercial lager beer Sensory evaluation score sheet

Number of panelist	Colour	Taste	Aroma	Mouthfeel	Appearance	Overall acceptability
1	8.97	7.93	7.73	8.69	8.87	8.44
2	8.83	8.07	7.87	8.79	8.73	8.46
3	9.21	7.69	7.49	8.98	9.11	8.50
4	8.95	7.95	7.75	8.67	8.85	8.43
5	8.94	7.96	7.76	8.68	8.84	8.44
5	9.02	7.88	7.68	8.69	8.92	8.44
6	8.93	7.97	7.77	8.85	8.83	8.47
7	9.27	7.63	7.43	8.88	9.17	8.48
8	8.81	8.09	7.89	8.85	8.71	8.47
9	8.85	8.05	7.85	8.66	8.75	8.43
10	8.92	7.98	7.78	8.69	8.82	8.44
11	8.51	8.39	8.27	8.31	8.41	8.38
12	8.41	8.49	8.13	8.21	8.31	8.31
13	8.22	8.68	8.51	8.02	8.12	8.31
14	8.53	8.37	8.25	8.33	8.43	8.38
15	8.52	8.38	8.24	8.32	8.42	8.38
16	8.51	8.39	8.32	8.31	8.41	8.39
17	8.35	8.55	8.23	8.15	8.25	8.31
18	8.32	8.58	8.57	8.12	8.22	8.36
19	8.35	8.55	8.11	8.15	8.25	8.28
20	8.54	8.36	8.15	8.34	8.44	8.37