

REGULAR ARTICLE

# INFLUENCE OF MALTING ON THE COMPOSITION OF MILLET (PENNISETUM TYPHOIDEUM) GRAINS AND THE GROWTH OF RHODOTORULA RUBRA AND TORULOPSIS CANDIDA

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#### SUMMARY

A comparative study on the influence of malting on the composition of *Pennisetum typhoideum* grains and the growth of *Rhodotorula rubra* and *Torulopsis candida* was investigated. Result of proximate analysis indicated high crude protein content of 11.04% in three day malted grains (3DMG) which decreased with decrease in malting period. The contents of sodium, potassium, manganese and iron were equally higher in 3DMG. The unmalted grains had high ascorbic acid content of 7.50 mg/g which decreased markedly to 5.00 mg/g in all the malted grains. The cellular masses attained after culturing the yeast for 15 days was insignificant (P> 0.05). The biomass of *R. rubra* and *T. candida* in 3DMG was 369 and 357 mg respectively. Comparatively, the biomass obtained in yeast dextrose peptone media (YDPM) was comparatively lower with 367 and 349 mg in R. rubra and T. candida. The result of this study suggest that malted P. typhoideum grains media could be used as an alternative medium for culturing *R. rubra* and *T. candida* in the laboratory against conventional expensive YDPM and 3DMG proved to be promising.

Keywords: Malting, Pennisetum typhoideum, Rhodotorula rubra, Torulopsis candida, Biomass.

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### 1. Introduction

Millet, (*Pennisetum typhoideum* (Burm) Stapf et C E Hubb) is one of the widely cultivated crops in Nigeria and in savannah regions immediately south of sahara. It is used as a cereal food, and in the production of locally brewed alcoholic beverages [1-3]. Chemical analysis of its grains showed that it contains carbohydrate, iron, phosphorus, calcium, vitamin B-complex and ascorbic acid [4]. The malted grains are commonly used for microbial growth and a number of culture media have been prepared from natural sources aimed at developing in amylases and proteases in the grains [5], and help to break down protein into forms which can be utilised by yeast [6]. Malting develop the enzymes that are required to modify the grain's starches into sugars [7].

Yeast is an essential ingredient in many bakery products responsible for leavening the dough and imparting delicious yeast fermenting flavour to the product [8]. It is used as food and also serves as a source of vitamins, especially B-vitamins which can be prescribed to patients with deficiency of B-vitamin [9]. Yeast can also be used for feeding animals and in brewing industries for alcohol production [10, 11]. In view of the importance attached to yeast, it is necessary to find alternative medium for culturing it other than the conventional yeasts dextrose peptone used in the laboratories, which at the moment is imported into the country and is expensive. Millet could be a better alternative due to its availability and affordability in the study area, cultivated predominantly in Sokoto and most parts of northern Nigeria. Despite the availability and the nutrient content of P. typhoideum, its potential as a source of medium for culturing yeast has not been investigated. This study reports on the influence of malting on the composition of *P. typhoideum* grains and its effect on the growth of Rhodotorula rubra and Torulopsis candida.

# 2. Materials and methods

#### Source of grains and malting process

Grains of millet (P. typhoideum) were purchased from the Sokoto central market, cleaned of dirt, stones, washed and dried. The dried grains were divided into four equal parts and three parts were separately thinly spread on moist jute sacks, and wetted with water. On the first notice of germination, one of the packs was harvested as one day malted millet grains, the rest two were harvested as a day, two days days malted millet grains and three respectively. The malted grains were sun dried by spreading out on clean mats. The un-malted millet grains sample and the three-malted millet grains sample were grinded separately into flour. They were packed in airtight polythene bags and kept at 35 ± 20C until needed.

#### Proximate and mineral analysis

Crude protein was estimated by Kjeldahl method as described [12]. Ash content was determined by incinerating 2g of the samples in muffle furnace and allowed to burn at 5500C [13]. Moisture was determined using the method described by [14]. Crude fat was extracted using Soxhlet extraction procedures [13]. The content of fibre, minerals and ascorbic acid content of the samples were analyzed in accordance with [15] method and absorbance taken using JENWAY 6100 spectrophotometer.

# Incorporation of the grains into a culture media

Two hundred grams (200g) of the unmalted millet grains was weighed and mixed with 200ml distilled water and then made up to 1000 ml. The mixture was brought to boil over a Bunsen burner flame for 20 min. and sieved with muslin cloth, the filtrate serving as broth medium of the un-malted millet grains. The malted grains were similarly prepared into media to obtain 1, 2 and 3 days malted millet medium respectively, and each of the medium was divided into aliquots of 49 ml and autoclaved.

#### Preparation of yeast extracts culture medium

The culture medium was constituted using 10 g of peptone, 5 g of yeast extract, 20 g of dextrose and 100 ml of distilled water. The peptone, the yeast extract, and the dextrose were first dissolved in 200 ml of distilled water by heating and stirring to ensure that the mixture had dissolved completely. The solution was made up to 1000 ml in volumetric flasks and autoclaved at 1210C for 15 min.

# Yeast starter culture inoculation of the millet media

Starter cultures of *Rhodotorula rubra* and *Torulopsis candida* were developed with yeast

stock obtained from Mycology Laboratory of the Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto. One ml each of the starter cultures was aseptically inoculated singly in a conical flask containing the un-malted, malted and the yeast extract culture media. The flasks were incubated on a culture shaker to provide adequate oxygen supply in the medium and to suppress fermentation and yeast growth was observed for 15 days.

### Measurement of growth in the yeasts

Yeast cultures were harvested at interval of three days and the growth of yeast cells was determined by the dry weight method. The yeasts produced in the medium were filtered with filter paper. Filtration took place until all the liquid content was filtered out living the yeast cell mass. Pouring distilled water into it through the filter paper washed the mass and the water was allowed to drain. The cell mass was dried in an oven at 700C to constant and weight determined. After drying, the weight of the yeast mass was obtained by subtracting the weight of the filter paper. The difference was recorded as the actual weight of the yeast.

# Statistical analysis

The result of this study was analysed using Pearson's  $X_2$  distribution and level of significance where necessary was considered at 5% level of probability.

# 3. Results and Discussion

The result of proximate analysis of malted and un-malted *P. typhoideum* grains evaluated is presented in Table 1. The 3 days malted millet and the 1 day malted millet samples had the highest moisture content of 10 % each, while, the un-malted and two days malted grains had 5 % moisture. Highest percentage

crude protein of 11.04 % was obtained in 3DMG followed by the un-malted and 2 days malted with 10.46 and 10.24 % respectively. The lowest value of protein (8.18 %) was obtained in 1 day malted. However, starch and protein digestibility values of millet at 48 h germination were significantly (P < 0.05) lower than samples germinated for longer period [16], which contradict the result of this study. The lowest fat content of 5.0 % was found in 3DMG while, the other three samples had 10.0%. Ash and fibre were traced in all the four millet samples, except the un-malted that had 5 % ash content. In this study, it was observed that malting decrease ash and fibre content of the grains.

The result of mineral and ascorbic acid analysis of P. typhoideum grains studied is depicted in Table 2. The content of sodium in 3DMG was high (5.65 mg/g), followed by two days and the un-malted with 4.56 mg/g each. The largest quantity of calcium (3.80 mg/g)was recorded in the un-malted millet sample and decreased to 2.50 mg/g in malted samples. The highest content of magnesium (21.3 mg/g)was observed in One-day malted millet and the un-malted with 12.7 mg/g. Magnesium content decreased with increase in malting period to 11.0 and 10.0 mg/g in 2 and 3DMG respectively. The un-malted millet sample had high content of phosphorus (3.5 mg/g), followed by 3DMG with 3.0 mg/g and one and two days malted had low content of 1.5 mg/g. The manganese content was highest in 3DMG (2.0 mg/g) and decreased to 1.4 and 1.3 mg/g in one and two days malted respectively. Similarly, high quantity of iron (10.6 mg/g)was obtained in 3DMG followed by un-malted millet with 7.60 mg/g which decrease in malting period to 4.70 mg/g in one and two days malted. The content of vitamin C was high in the un-malted samples with 7.50 mg/g,

while, the other three ma	ilted millet samples
had vitamin C of 5.0 m	g/g. In this study,

malting decreases ascorbic acid content of the grains.

Table 1: Proximate composition of malted and unmalted *P. typhoides* samples %.

Samples	Moisture	Ash	C/P	Fat	Fibre			
Un-malted millet	05.0	05.0	10.46	10.0	Trace			
One day malted millet	10.0	Trace	8.18	10.0	Trace			
Two days malted millet	05.0	Trace	10.24	10.0	Trace			
Three days malted millet	10.0	Trace	11.04	05.0	Trace			
CP = Crude  protein.								

Table 2: Ascorbic acid and minera	al content of millet samples	(mg/g).
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Sample	Vit. C	Na	К	Ca	Mg	Р	Mn	Fe
Un-malted millet	7.5	4.56	799.2	3.8	12.7	3.5	1.8	7.6
One-day malted millet Two-day malted millet	5.0 5.0	4.35 4.56	767.3 799.2	2.5 2.5	21.3 11 3	1.5 1.5	1.4 1 3	4.7 4.7
Three-day malted millet		4.50 5.65	1023.0	2.5	10.0	3.0	2.0	10.6

Each value represents mean of three replicates.

The growth of *R. rubra* in 3DMG and YDPM was insignificant (P>0.05) different (Table 3). However, there is marked difference between the biomass of R. rubra in 3DMG and YDPM with 369 and 367 mg respectively. Generally, the malted millet medium was more supportive on the growth of the yeast than the un-malted millet medium and 3DMG had the highest, followed by two and one- day

malted medium (Table 3). Similarly, the growth of *T. candida* was highest on the 3DMG (357 mg), followed by two and one-day malted millet medium with 278 and 212 mg respectively. Growth on 3DMG is comparable with growth on YDPM (349 mg). There was no notable difference between the mean growths obtained by *T. candida* in the two media (Table 4).

Table 3: Biomass (mg) of *R. rubra* on different types of broth media for 15 days.

Broth media		sampling period (days)							
	3	6	9	12	15	Mean	+ S.E		
Un-malted millet		158	169	239	245	246	211	19.67	
One-day malted millet		160	185	257	266	252	224	21.51	
Two-day malted millet.		167	226	253	375	373	299	42.97	
Three-day malted millet.		269	359	378	424	413	369	27.52	
Yeast dextrose peptone		270	354	367	422	424	367	28.15	

X2 = 36.877, S.E = Standard Error.

3	6	9	12	15	Mean	+ S.E	
	135	153	221	236	237	196	27.76
	143	176	241	251	250	212	17.47
	148	200	342	350	349	278	43.19
	239	347	365	423	412	357	32.76
	235	344	359	421	422	349	34.36
	-	135 143 148 . 239 235	135      153        143      176        148      200        239      347	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	3      6      9      12      15      Mean      + S.E        135      153      221      236      237      196        143      176      241      251      250      212        148      200      342      350      349      278        239      347      365      423      412      357

Table 4: Biomass (mg) of *T. candida* on different broth media for 15 days.

X2 = 36.585, S.E = Standard Error.

Compositionally, the 3DMG had high percentage of crude protein, sodium, potassium, phosphorus, manganese, vitamins, and iron while, YDPM composed primarily of peptone as a source of amino acids, nitrogen, sulphur and phosphorus; yeast extract as a source of vitamins, and other growth factors; and the dextrose as the source of carbon and energy. The higher growth observed in 3DMG is comparable to the yield in YDPM. The declined growth observed in two and one-day malted millet media as compared with 3DMG that supported higher yields, suggest that nutrients are more available for the yeasts to utilize in 3DMG. The higher yield observed may be due to the presence of protein, as amino acids

#### REFERENCES

- 1. Oyenuga, V.A. (1968) Nigeria's foods and feeding stuff 3rd edn. Ibadan. University Press, Ibadan, Nigeria.
- Opoku, A.R. Ohenhen, S.O. and Ejiofor N. (1981). Nutrient composition of millet (*Pennisetum typhoides*) grains and malt. J. Agric. Food Chem. 29(6): 1247-1248.
- Iwuagwu, Y.O.U. and Izuagbe, Y.S. (2008). Studies on the preservation and bottling of oyokpo-a Nigerian beer from millet (*Pennisetum typhoideum*). Journal of Applied Microbiology 59(6): 487 - 492
- 4. Babu, B.V., Ranmana, T. and Radhakrishna, T. M. (1987). Chemical

play important role in successful growth and fermentation since they are the building blocks of essential proteins [17]. In addition, 3DMG and YDPM both have phosphorus, sulphur, and manganese which are essential for the metabolism of yeasts. According to [18, 19], manganese plays a very vital role in the metabolism of yeast and supports protein and thiamine synthesis. The result of this study suggest that malted *P. typhoideum* grains media could be used as alternative medium for culturing *R. rubra* and *T. candida* in the laboratory against conventional expensive YDPM and, 3DMG proved to be promising.

composition and protein content in hybrid varieties of finger millet. Indian J. Agric. Sci. 57:520-522.

- Okafor, N. (1985). Industrial Microbiology. Univ. Press, Ife. 2nd edition pp 174 – 255.
- 6. Briggs, D. E. (1998). Malt and Malting, Kluwer Academic/Plenum publishers.
- 7. Clark, C. (1998). The British Malting Industry Since 1830, Hambledon Continuum.
- McGee, H. (1988). On Food and Cooking. Collins 2nd Edition, New York. Pp 306 - 311.
- 9. Edozien, J.C. (1969). Yeast for Human Feeding; New Data on Safety. FAO/WHO/UNICEFProtein Advisory

Group, United Nations, PAG Document 2/23/1 (United Nations, New York)

- 10. Butt, K.R. (1993). Utilisation of solid paper-mill sludge and spent brewery yeast as a feed for soil-dwelling earthworms. Bioresource Technoloy. 44(2): 105-107.
- 11. Waites, M. J. Morgan, N.L., Rockey, J.S. and Higton, G. (2001). Industrial Microbiology: An Introduction. Wiley-Blackwell, Canada. 304 pp.
- Bakare, B. (1988). Method of Biochemical Analysis of Plant Tissue. Agronomy Department, Univ. of Ibadan.
- 13. Pearson, D. (1976). The Chemical Analysis of Food (7th ed.). Churchill Livingstone, London pp. 6-25.
- 14. Udo, E.J., and Oguwale J.A. (1986). Lab. Manual for the Analsysis of Soil, plant and water samples. 2nd ed. Uni. Press Plc. Pp. 147-150.
- Association of Official Analytical Chemistry. (1990). Official Methods in Analysis. 15th edition. Published by Association of Official Analytical Chemist Inc. pp 241 – 248.
- Chaturvedi, A. Sarojini, G. (1996). Malting of pearl millet (*Pennisetum typhoideum*): Its effect on starch and protein digestibilities. Journal of food science and technology 33(4): 342-344.
- Ostergaard, S., Olsson, L., and Nielson, J. (2000). Metabolic engineering of *Saccharomyces cerevisiae*. Microbiol. Rev. 64(1): 34-50.
- Keeton, W.T. (1976). Biological Science.
  3rd Edition W.W. Norton and Co. New York pp 252 – 255.
- Carveny, K.L., McCaffery, J.M., Jensen, R.E. (2001). Division of mitochondria requires a novel DMNI Interacting protein, Net 2p. Mol. Biol. Cell. 12(2): 309 – 321.