

APPLICATION OF CHEMICAL TREATMENT TO ENHANCEMENT OF FUNCTIONAL PROPERTIES OF WHEAT GLUTEN

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Abstract: The present study was carried out with the aim to enhance the functional and rheological properties of the gluten from Chandousi Wheat variety by the L-Ascorbic Acid Chemical treatment. During project work, wheat flour of the Chandousi variety was kneaded to dough by adding the L-Ascorbic acid solution in ratio 20 ppm to 120 ppm (20 ppm, 40 ppm, 60 ppm, 80 ppm, 100 ppm and 120 ppm) the dough was submerged in the distilled water for one hour and then the dough was placed under the constant stream of water to isolate the gluten. The achieved gluten was freeze dried and was evaluated for various physicochemical properties. The result shown the optimum dose to be 60 ppm on the basis of the improvement in the water absorption capacity, sedimentation value, foaming capacity and emulsifying capacity, compared with non-modified gluten powder (control) Modified and non-modified gluten powder were then integrated in the noodles base dough of Lokvan variety of wheat flour. FE-SEM images of non-modified gluten and chemical modified freeze dried gluten were taken for identification and conformation of the modification on morphological basis. Dough obtained and the noodles dough were compared on the basis of Brabender's Farinograph and Extensograph. The result concluded improvement in the noodles dough as well noodles made from the chemical treated gluten as compared to control sample.

Key words: *Wheat, Wheat flour, Chandousi, Lokvan, Noodles.*

1. INTRODUCTION

Gluten is a protein macro-polymer in wheat flour which is a mixture of more than 100 heterogeneous polypeptides, that are formed in hydrated flour during dough mixing and Gluten is composed of two main storage proteins, namely, Gliadins and glutenins. Gluten plays a major role in viscoelastic properties of bread-making which is highly correlated to the quality of end products. Glutenins and gliadins are the polymeric and monomeric protein components of gluten, respectively. Glutenins (with molecular mass of 69 to 88 kDa based on SDS-PAGE) are responsible for elastic behavior, whereas gliadins (with molecular mass of 30 to 50 kDa) are responsible for viscous flow properties of the foods.

Wheat flour is the most important ingredient in the bakery industry. It provides bulk and structure to most of the baker's products, including breads, cakes, cookies, and pastries. While the home cook depends almost entirely on a product called all-purpose flour, the professional baker has available a wide variety of flours with different qualities and characteristics. In order to select the proper flour for each product and to handle each correctly, we should understand each type of flour and how it is milled (Gupta, et al. 1993).

Bran and germ proteins have a higher content of essential amino acids than the inner endosperm. In the wheat endosperm, a prolamine (called gliadin) and a glutelin (called glutenin) are present in approximately the same concentrations; in the bran a prolamin is most abundant with fair amounts of an albumin and globulin. The unique presence of glutenin and gliadin in the wheat endosperm is important to the baking operation. In the presence of water and with mechanical agitation, these protein fractions form a tough, elastic complex termed gluten which is capable of retaining gases and by doing so makes a leavened product possible (Huebner, et al., 1997).

Proteins have many useful functional properties in foods. A functional property is a characteristic of the protein that enables it to perform a specific role or function in a food. For example, a protein with the ability to form a gel may be used in a food with the specific intention of forming a gel, as in use of gelatin to make jelly. Functional properties or roles of proteins in foods include solubility and nutritional value. They also may be used as thickening, binding, or gelling agents and as emulsifiers or foaming agents. (Graybosch et al., 1996). The ability of wheat gluten to form a visco elastic mass when fully hydrated sets it apart from all other commercially available vegetable proteins. Gluten has gained wide acceptance in the food processing industries because of its unique physical properties, such as visco-elasticity, film-forming ability, thermosetting properties and high water absorption capacity. Its visco-elastic properties improve dough strength, mixing tolerance and handling properties in a bread making process. The film forming property of hydrated gluten is a result of its elasticity the film forming ability of gluten enhances gas retention and controlled expansion for improved volume, uniformity and texture of baked products (Kaushik R. et. all. 2004).

2. MATERIALS AND METHODS

Raw Material: For this research Chandousi wheat and Lokwan wheat these local varieties were purchased from the local market, Navjeevan super market, Jalgaon.

Chemicals and glassware's: The Foodgrade Standard chemicals brand (SDFCL, MERCK,) were obtained from local chemicals supplier, Kishore Agency, New B.J. Market, Jalgaon.

Equipment's and machineries: The equipment and machineries like Soxhlet apparatus, Kjeldhal, muffle furnace and hot air oven. Hot Air Oven (Lab Hosp) Freeze dryer, FE-SEM- Field Emission- Scanning Electron

Microscopy, Brabenders Farinograph Unit, Extensograph, Centrifuge equipment was used for the present investigation were used from the Food Engineering, Jalgaon.

2.1 Preparation of Dough and L Ascorbic acid Treatment :

The dough was prepared by method described by American Association of Cereal Chemists (AACC) and gluten was isolated by hand washing method as detailed in AACC (2000a) method No. 38-10 (AACC). Following is the method for non-modified and freeze dried gluten powder.

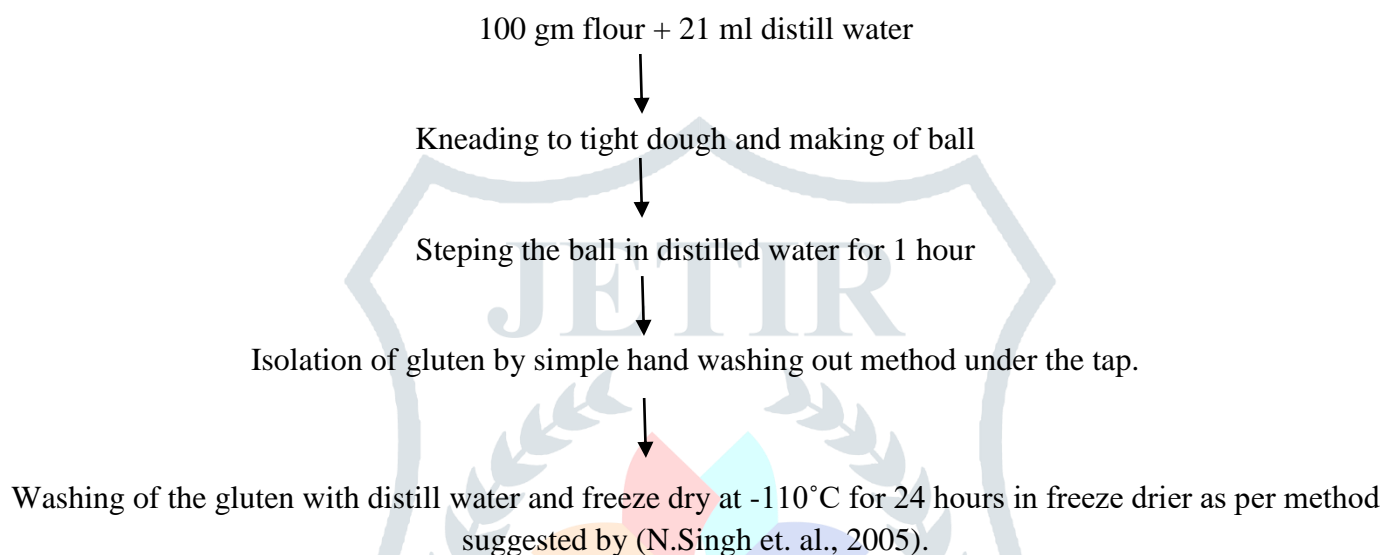


Figure No. 1. Preparation of dough and L-ascorbic acid treatment

2.2 Isolation of Gluten and its Freeze Drying

Method for L-ascorbic acid treatment and modification was followed and obtained from (V. Kolpakova et. al., 2014) with some minor modification by changing the Chemicals and concentration in ppm level. 100 gm of the Chandusi and lokwan Wheat flour was kneaded by adding the 5 ppm concentrated l ascorbic acid solution of 28 ml, after kneading it was kept immersed in the water for 1 hour. After 1 hour, the gluten was isolated by the hand washing method. The similar process was repeated for the every concentration from 20 ppm to 120 ppm. On the completion of isolation of gluten from the treated dough which was modified by the L-ascorbic acid and gluten was freeze dried in (Lypolizer- SCANVAS- SANSKAM Technologies Pvt. Ltd.). The by-products such as bran; germ and starch mixture and pure starch were recovered.

2.3 Method for preparation of noodles from treated wheat flour

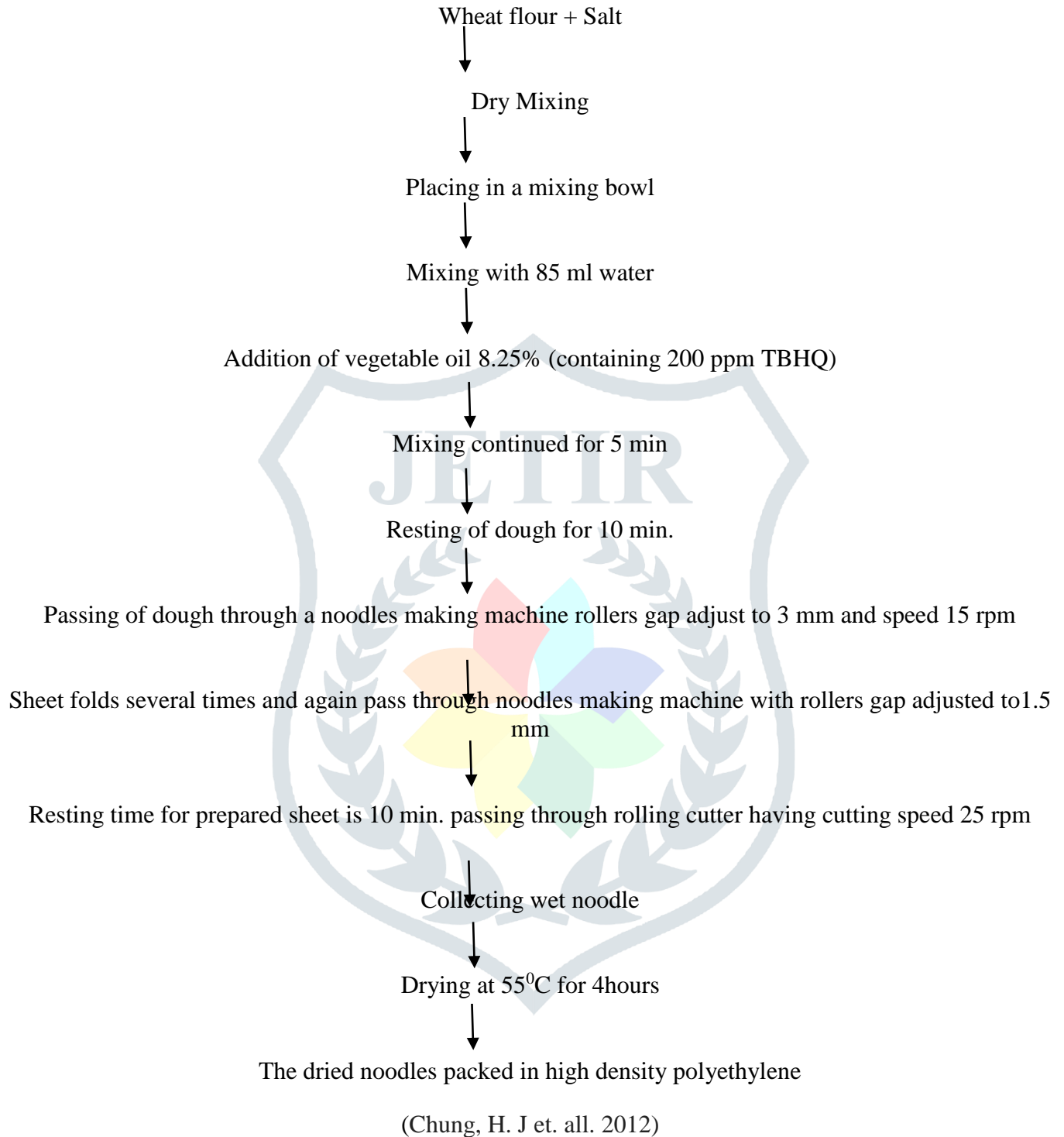


Figure No. 2. Flow sheet for Preparation of Control Noodles

2.4 Characterization and isolation of Treated gluten and non-treated gluten

2.4.1 Proximate analysis: The proximate compositions of wheat flour and crude protein, crude fat, crude ash and crude fibre estimation of gluten and functional properties were determined by approved methods 44-40, 46-11A, 30-10, 08-01 and 32-07, respectively (AACC, 2000). After the analysing the physicochemical

parameters of the wheat grains the grains were converted to flours at the local flour mill, located at Bhambhori, Jalgaon.

2.2.2 Sensory evaluation: The nine-point hedonic scale was used for conducting the sensory evaluation of noodles (Joshi, V. K., 2006) by using 10 semi-trained judges (age around 24-50 year) i.e. faculty members and research students of the North Maharashtra University Jalgaon. The panellists were asked to evaluate the various samples for different sensory attributes namely appearance, colour, texture, flavour and overall acceptability. The sample was presented to judge and plain water was given to the rinse their mouth in between the evaluation of the sample. Sensory evaluation was done at $32\pm^{\circ}\text{C}$, and the mean score of all attributes was used to draw overall acceptability of the product (Kumar, V et. all 2018)

2.2.3 Statistical analysis: Each manufacturing treatment and physico-chemical measurement was carried out in triplicates. The experimental data were subjected to statistical evaluation using analysis of variance (ANOVA) for a completely random design using Statistical Analysis System. Duncan's multiple range tests were used to determine the difference among means and the significance was defined at $P < 0.05$. (Frostegard, A. et, all. 1993)

3. Results and Discussion

The Lokwan Wheat variety had the longest grain length of 4.911 mm and Chandousi had lower grain length of 4.6mm. Chandousi thousand kernel weight of 40.12 gm. The thousand kernel weight of the Lokwan Wheat was highest of 50.06 gm. The Chandousi Wheat had the smallest grain length but has good thousand and single kernel weight. The sieve residue showed the amount of damaged, broken and under sized kernel, it should be minimum to avoid the losses. The Lokwan had smallest amount of sieve residue while Chandousi had the highest. It concludes that chhandousi had more damaged and under sized kernels (Food Safety Standards Regulations, 2011)

The Lokwan was of greater order. The angle of repose shows the flow ability of the grains (Khazaei J. and Ghanbari S. 2010). The minimum angle of repose gives excellent flow ability. Lokwan had the minimum angle of repose of 25.11° ; hence it showed the excellent flow ability. The Chandousi Wheat had 26.06° Following table 1 is showing the results for physicochemical analysis of two wheat varieties Lokwan Whole Wheat, Chandousi Whole Wheat. Lokwan varieties of wheat found abundantly in Maharashtra.

3.1 Physicochemical Parameter of Whole Wheat Grain:

The bulk density of the Chandousi Wheat was highest and lokwan wheat had lowest. The moisture content of Chandousi wheat was highest and lokwan have lowest one. All the moisture contents of whole wheat grain

were within the limits which were prescribed by the (ISO 1666 and Food Safety Standards Regulations, 2011; moisture not more than 14 %). From all the results of the Physicochemical parameter which were analyzed in the laboratory for the whole wheat grains of all the two varieties, the lokwan was of lower physical parameter and Chandousi had higher physical parameter, which concludes that quality of lokwan was low and quality of chandousi was high., considering the values that obtained after analysis. The angle of repose for the Lokwan and Chandousi Wheat Flour was of 45° while 40° for the chandousi Wheat Flour. The sedimentation and dispersibility are the inversely proportional terms. When sedimentation is lower the dispersibility is higher and vice-versa. The sedimentation value of the chandousi Wheat flour was highest 38.1mL, showing the good quality protein and Lokwan Wheat Flour had 29.1 mL of sedimentation value respectively.

Table 1. Results of Physicochemical Parameter for Whole Wheat Grains

Sr .No.	Parameter	Chandousi	Lokwan
1	Single kernel weight(gm)	0.06± 0.09	0.04 ± 0.06
2	Grain Length (m	4.6 ± 0.04	4.9 ± 0.6
3	1000 Kernel Weight (gm)	40.12 ± 0.17	50.06 ± 0.25
4	Angle of Repose (0)	26.06 ± 0.05	25.11 ± 0.22
5	Sieve Residue (gm)	160.15± 0.35	148.86 ± 1.24
6	Moisture (%)	7.12 ± 0	6.45 ± 0.01

All values are means of three determinations ± standard deviation (SD)

The moisture of all the two flours was within the limit as prescribed by FSSAI (less than 14 %). The ash content and the acid insoluble ash of all the two wheat flours were in prescribed limit of less than 2.5 and 0.15% respectively (Food Safety Standards Regulations, 2011). The crude fibers in all the three wheat flours were within the prescribed (Food Safety Standards Regulations, 2011)

3.2 Physicochemical Analysis of Wheat Flours:

The Chandousi Wheat Flour had the highest amount of crude protein of 12.03% while, Lokwan had the lower crude protein 8.64 % respectively. Table No. 2 indicates that the gluten content of the Lokwan was 27.4. % (on wet basis) and 9.54 % (on dry basis). While the chandousi Wheat had highest gluten content of 30.53 % (on wet basis) and 10.5 % (on dry basis) respectively. Hence the chandousi Wheat gluten was selected to isolate and modify.

Table 2 Results of the Physicochemical Analysis of Wheat Flours

Sr No	Parameter	Chandousi	Lokwan
1	Angle of Repose (gm/ml)	40 ± 0	45± 0
2	Bulk Density	0.70 ± 0.01	0.65± 0.05
3	Moisture (%)	8.31± 0.20	7.35± 0.08
4	Dispersibility (%)	68.3 ± 0.56	57.7± 0.68
5	Sedimentation (ml)	38.1±1	29.1± 0.25
6	Ash (%)	0.79±0.01	0.48± .0.01
7	Acid Insoluble Ash (%)	1.01±0.01	.0.85± 0.05
8	Crude Fibre	0.19± 0.01	0.49± 0.03
9	Protein (%)	12.03± 0.33	8.64± 0.51
10	Gluten wet basis (%)	30.53±0.87	27.4± 0.87
11	Gluten Dry basis (%)	10.5± 0.51	9.12± 0.58
12	Solvent Retention Capacity (%)		
12.1	Deionized water	135.3± 0.76	153.5± 1.14
12.2	5% Sodium carbonate	351.7± 0.85	236.1±0.94
12.3	5% Lactic Acid	271.4± 1.05	239± 0.98
12.4	50% sucrose	365.1± 2.07	295.5± 1.28
13	Solubility	10.8±0.15	8.94± 0.08
14	Water Absorption capacity	255.5± 1.54	225.5± 0.42
15	Foaming Volume (ml)	15.3 ± 0.50	12.2± 0.72

All values are means of three determinations ± standard deviation (SD)

Solvent Retention Capacities for the water and 50% sucrose solution. The Chandusi Wheat flour had the best SRC for all the solvent except the Sodium Carbonate, as it gave the number of damaged starch proportion. The water solubility index for the chandousi Wheat flour was highest of 13.59 and Lokwan, Wheat flour had the lowest respectively. The water solubility index shows up to a degree of the flour that is soluble in water. While water absorption capacity was calculated in percent in which chandousi Wheat flour and Lokwan wheat flour had 255.5% and 225.5% respectively the foam volume shown by the 1 gm of the chandousi Wheat flour was 15 mL while the Lokwan gave lower foam volume of 12.2 mL respectively.

The table No. 3 is showing the results for the freeze dried gluten powder isolated from the chandousi wheat without any treatment, which was subjected to further treatments, viz. chemical and enzymatic. The bulk density of the freeze dried gluten powder was 0.71. The results of the chemical analysis of the freeze dried gluten powder were all in limits which were prescribed by (Food Safety Standards Regulations, 2011). The ash content, moisture content and acid insoluble ash content of the freeze dried gluten powder were in the prescribed limit (Food Safety Standards Regulations, 2011 and ISO 1666 and ISO 3593). The dispersibility was 68.6 % and the sedimentation value was 29.1 mL. The water absorption capacity was 1.26 %. The oil holding capacity was of 1.89 %. The oil holding capacity and water holding capacity of the gluten powder was same as prescribed (Janica Charelle S. Borja, 2013). It showed good emulsification capacity of 9.33 %. The foaming volume was of 6.6 mL. The gluten powder showed

Table 3 Results of the Physicochemical Analysis of Non-Modified Freeze Dried

Gluten of chandousi Wheat Sr.no	Parameter	Result
1	Moisture	6.89± 0.05
2	Ash	0.58± 0.10
3	Acid Insoluble Ash	0.097±0.30
4	Bulk Density	0.71 ±0.05
5	Dispersibility	68.6± 0.15
6	Sedimentation	29.1± 0.10
7	Water Absorption	1.26±0.01
8	Oil Absorption Capacity	1.89± 0.08
9	Foaming Capacity	6.66± 0.57
10	Emulsification Capacity	9.03 ± 0.09
11	Solubility	3.04 ± 0.08
12	Solvent Retention Capacity %	
13	Deionized Water	231.9 ±0.15
14	50% sucrose	292.6±0.67
15	5% Lactic Acid	220.±0.14
16	5% Sodium carbonate	194.8± 0.15

All values are means of three determinations ± standard deviation (SD)

292.6 % of Sucrose retention and 194.8 % of sodium carbonates retention. The lactic acid retention was 220 %, which showed the gluten was of good quality.

Table 4 Results of the Physicochemical Analysis of Ascorbic Acid Modified Gluten

Sr no.	Parameter	GA1	GA2	GA3	GA4	GA5	GA 6
1	Moisture %	7.10±0.1	6.9±0.0	7.32±0.1	7.1±0.1	7.31±0.1	7.32±0.1
2	Ash %	0.61±0.01	0.63±0.40	0.65±0.01	0.63±0.50	0.64±0.01	0.65±0.35
3	Acid Insoluble Ash%	0.088±0.0	0.083±0.05	0.079±0.01	0.074±0.05	0.083±0.03	0.082±0.02
4	Bulk Density (ml)	0.67±0.01	0.68±0.05	0.67±0.05	0.67±0.05	0.67±0.01	0.68±0.01
5	Dispersibility %	67±1	67.6±0.57	68.1±0.57	67.6±0.57	67±1	65.6±0.57
6	Sedimentation	31.3±1.01	29.6±0.57	33.6±0.57	29.6±0.05	29.6±0.57	30.6±1.01
7	Water Absorption Capacity	2.67±0.91	2.46±0.08	2.78±1.09	2.56±0.35	2.40±1.10	2.46±1.14
8	Oil Binding Capacity	1.48±0.09	1.61±0.07	1.64±0.13	1.43±0.05	1.69±0.01	1.59±0.01
9	Emulsification Capacity	7.26±0.08	7.07±0.05	7.37±0.07	7.21±0.04	7.06±0.04	7.15±0.03
10	Foaming Capacity	7.60±0.10	6.93±0.15	7.83±0.15	6.98±0.67	6.9±0.1	6.91±0.12
11	Solubility	2.87±0.05	3.05±0.03	3.15±0.07	3.11±0.07	2.036±1.52	3.08±0.06
12	Solvent Retention Capacity						
13	Deionized Water	228.6±1.14	221.5±0.81	220.4±0.97	212±0.71	215±1.04	226.5±1.18
14	50% Sucrose	251.5±1.04	261.3±0.92	267.3±0.63	286.4±1.35	261.4±0.69	234±1.33
15	5% Lactic Acid	260.2±1.00	259.6±1.02	263.9±0.55	255.4±1.06	250.9±1.19	257.5±0.82
16	5% Sodium Carbonate	202.2±0.85	197.4±0.97	199.6±1.19	203.6±0.62	207.9±0.31	178.1±0.31

All values are means of three determinations \pm standard deviation (SD)

It remained as before the same and identical to the values before the chemical treatment and modification. The changes were found in the following parameters. The dispersibility value was decreased by 1% and the sedimentation was increased by 4 ml. the Sample Ga3 gave highest sedimentation value of 33.6 ml proving good quality protein. The oil holding capacity was slightly decreased by 0.25 % and the water absorption capacity was also increased. The water solubility was also affected and deviation was found in decreased order of difference showing the 1.02 %. The Solvent Retention Capacity was also affected by the L-Ascorbic acid. The Lactic acid retention was best shown by the Sample Ga3. It proved that the quality of the gluten protein was improved by the L-Ascorbic acid treatment with the dose of 60 ppm.

3.3 Results for Rheological Testing

3.3.1 Farinograph:

According to the analysis results of the modified and freeze dried gluten samples of Chemical modification (L-Ascorbic acid treatment) and enzymatic modification Papain (protease). From each of the treated sample one sample was selected for the further rheological testing's Farinograph, and Extensograph.

The table No.4 is showing the list of selected samples of modified gluten which are too incorporated in the flours of Lokwan Whole Wheat Flour. The samples were selected on the basis of the results of physicochemical analysis and the functional properties.

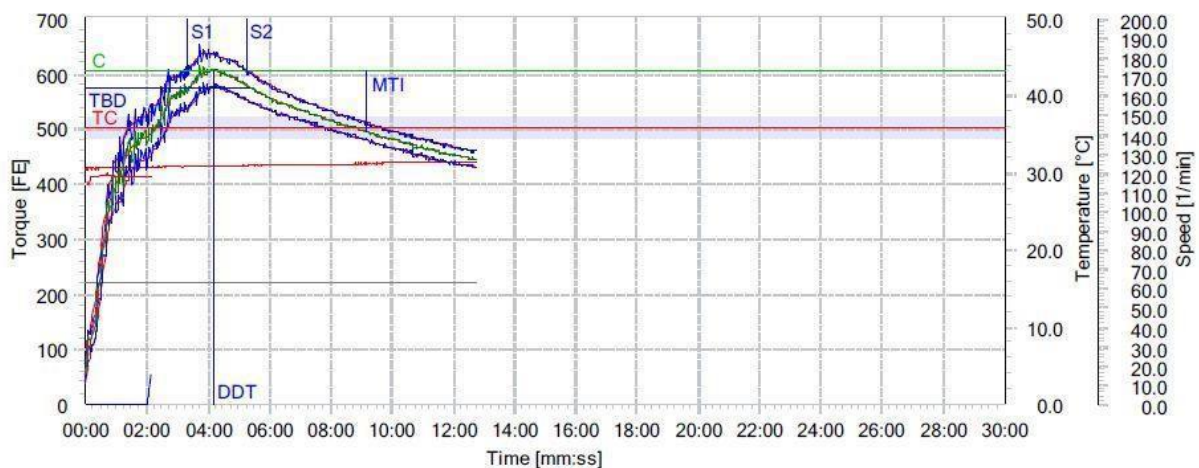


Fig. No. 3. Farinograph for Lokwan Whole Wheat Flour Fortified with Non -Modified Freeze Dried Gluten

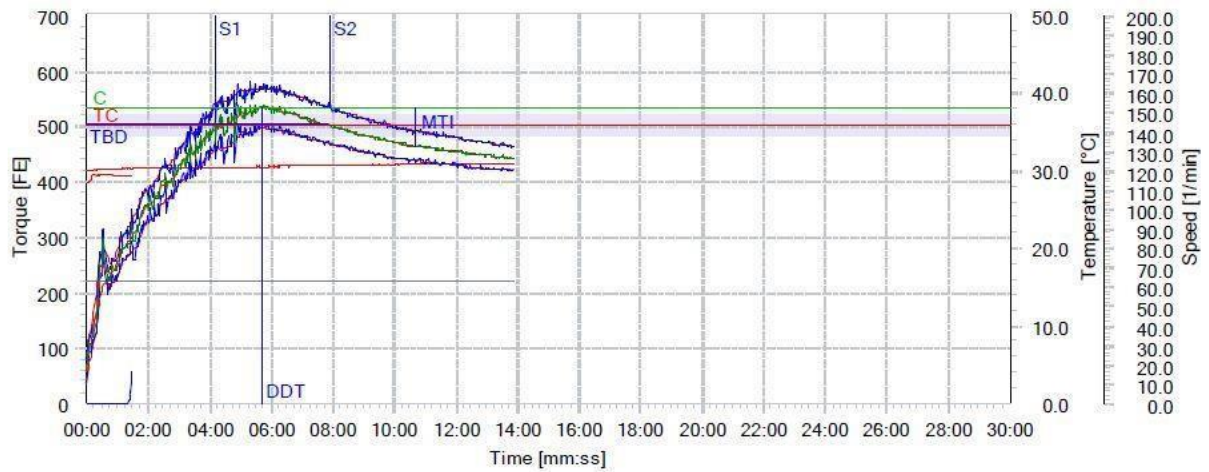


Figure No. 4. Farinograph for Lokwan Whole Wheat Flour Fortified with L- Ascorbic Acid Modified Gluten (Ga3)

Table No. 5. Farinograph Result - Mixing Characteristics of Dough (Lokwan

Sr. No.	Name of Dough	AT (min)	PT (min)	DT (min)	MTI (BU)
1	NMG	2.0	4.10	29.6	111
2	AAMG	1.7	5.44	29.3	70
3	DMG	2.1	5.27	28.6	44
4	PMG	1.8	4.44	28.2	62

As per above Farinograph data, adding modified gluten powder to Lokwan Whole Wheat flour produced the better effects than flour alone, meaning that mixing characteristics and gluten strength were improved; however, the farinograph data may not be an accurate representation for the effects of Papain (protease) on gluten strength. The poor results were similar to that observed by Tayyaba Naseem, strengthened dough, whereas, Gluten produced a higher peak time, had a lower weakening slope meaning that it was more tolerant to over mixing. The results for the L-Ascorbic Acid were best as compared to papain modified gluten. The flour with L- Ascorbic acid modified gluten had indicate the highest mixing tolerance index of 70BU. Datem modified gluten had indicate the lowest mixing tolerance index as compare to L- Ascorbic Acid modified gluten and papain modified gluten .

3.3.2 Extensograph:

Results for the rheological testing (Extensibility) of the Lokwan Whole Wheat flour fortified with L-Ascorbic Acid modified gluten powder is shown the Figures 5.

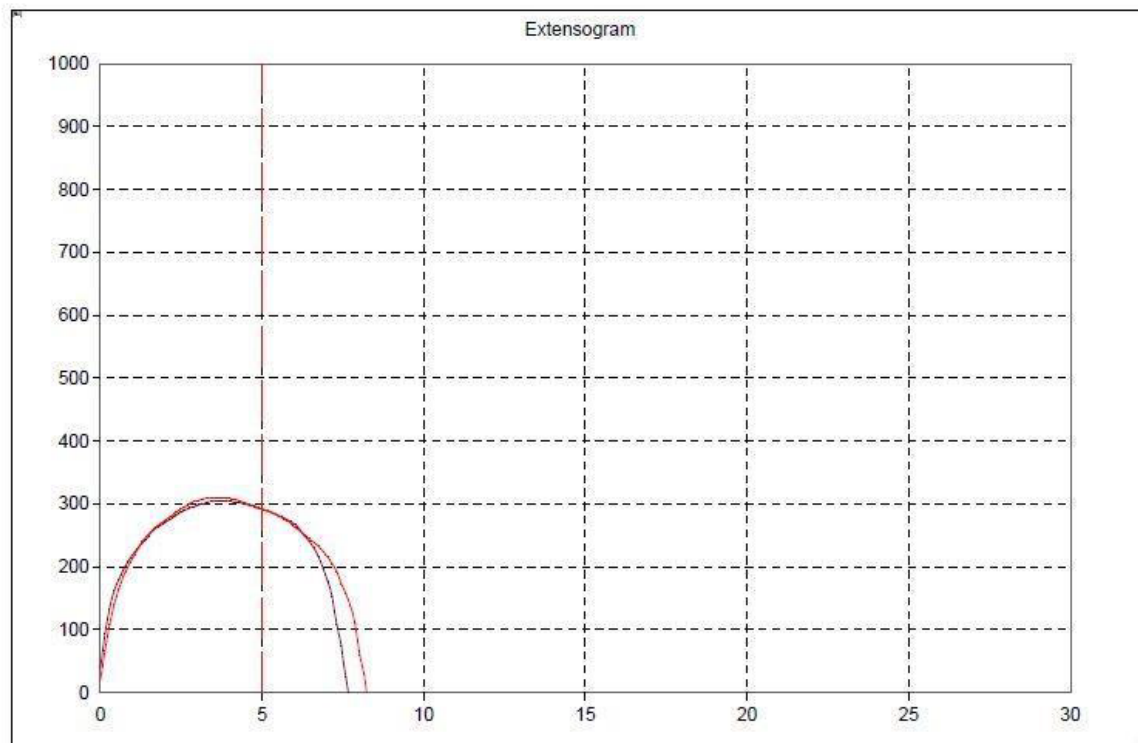
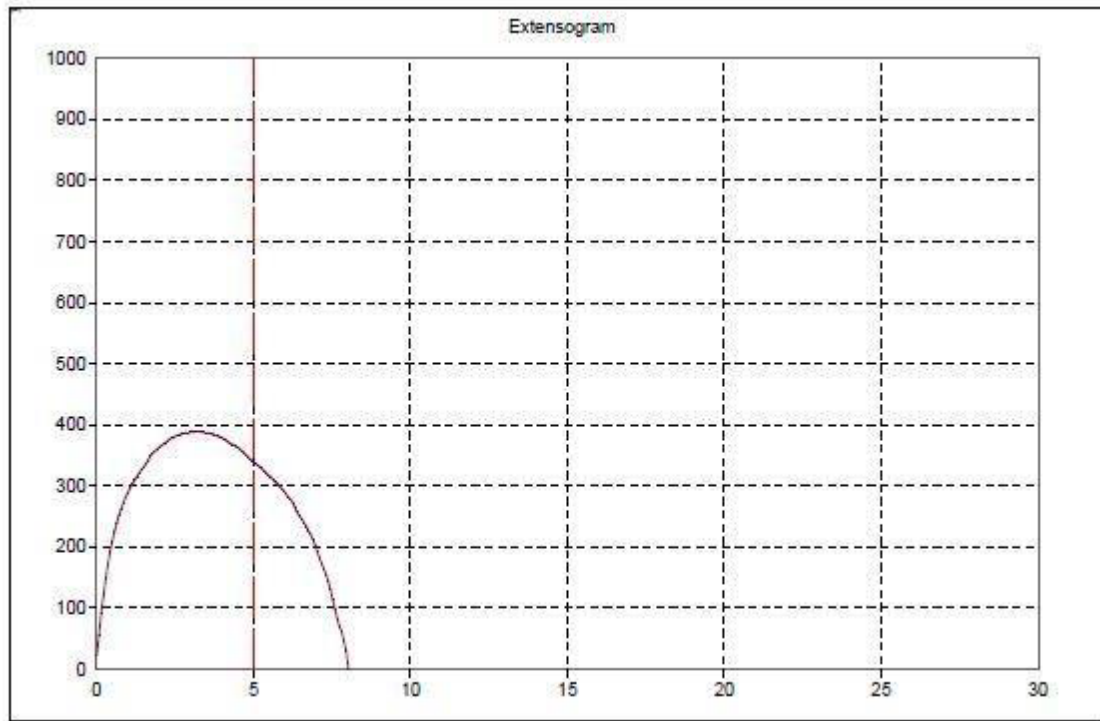


Figure No. 5 Extensograph for the Lokwan Whole Wheat Flour Fortified with L- Ascorbic acid Modified Gluten (Ga3)

Results for the rheological testing (Extensibility) of the Lokwan Whole Wheat flour fortified with gluten powder is shown the Figures 6



Test: C:\Users\Extensograph\Documents\Date+m+wheat flour-350.EXD

Figure No. 6 Extensograph for the Lokwan Whole Wheat Flour Fortified Modified Gluten (Ga3)

Table No. 6 Results of Extensograph for Fortified Lokwan Whole Wheat Flour

Sr. No.	Parameter	Non – Modified Gluten	L-Ascorbic Acid Modified
1	R	249	291
2	E	103.5	79.5
3	R/E	2.45	3.65

4. CONCLUSION

In contemporary work was carried out to Enhance the functional properties of Wheat gluten by implementing Chemical and Enzymatic treatment and further integrate the wheat gluten powder In to the wheat flour (LOKWAN) for the production of Noodles. The physical and chemical parameter of wheat flour (Chandousi) was evaluated. The Chandousi Wheat was observed highest gluten content as compared to LOKWAN wheat variety there for Chandousi was selected to modify and further to integrate into the LOKWAN wheat variety. The dough of the Chandousi was treated with the L-Ascorbic Acid, The gluten was basically extracted by the hand washing out method. Then the extracted gluten was freeze dried at -110 °C. The freeze dried and modified

gluten was characterized for the functional properties and the conformation of the modification was accomplished by incorporating the modified gluten into the Lokwan Whole Wheat flour. The rheological properties were estimated by Brabenders Farinograph, and Extensograph.

The addition of the modified gluten to the whole wheat flour (Lokwan) brought improvement in the dough extensibility, elasticity, Mixing Tolerance Index, dough gluten quality and quantity. It is possible to produce improved noodles from lokwan wheat variety by incorporating modified glutes obtained from chandousi wheat flour with the treatments 60 ppm of L-Ascorbic Acid, incorporation) however the modified gluten shown best result.

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