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Rheological and quality characteristics of thyme (Thymus vulgaris) enriched bread

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Abstract
In the present study, the effect of addition of dried thyme at 1, 2, 3 and 4% in wheat flour was examined to prepare functional bread. Addition of thyme significantly increased the water absorption rate and mixing tolerance index of flour. However, dough stability was decreased as the level of supplementation of thyme increased in the flour. Therefore, little modifications were done in baking procedure to prepare thyme bread. Presence of thyme in the formulation had a negative impact on the specific volume of bread. From sensory point of view, 2% level of thyme in the bread was selected as best. Thyme bread was high in total phenolic content and radical scavenging activity. Altogether, our findings indicate that thyme could be added as a functional ingredient in the bakery products.

Keywords: Thyme, rheology, total phenolic content, antioxidant activity, bread

Introduction
Thyme is rich in health promoting phytochemicals, fibre, minerals and vitamins. Thyme is very popular among culinary herb plants, originally native to southern Europe and Mediterranean regions. In the Mesopotamian cradle of civilization, humans were using thyme for its health promoting properties as early as 5,000 B.C. [6, 7, 9]. Ancient Greeks used thyme for incense while the Romans were known to add thyme to cheese and alcoholic beverages. The fresh or dried leaves of thyme were widely used to flavor soups, stews, baked or sauteed vegetables, casseroles and custards. Thyme essential oil is rich in thymol and carvacrol, possesses strong antiseptic, antibacterial and antioxidant properties. These terpenes bind to the amine and hydroxylamine groups of the proteins of the microbial membrane altering their permeability and resulting in the death of the microorganism [17, 5, 29, 10].
Scientist observed inhibitory action of thyme (*Thymus vulgaris*) on *S. aureus, S. typhimurium* and *V. parahaemolyticus*. Scientist investigated the effect of the thyme essential oils against *Escherichia coli, Listeria monocytogenes, Staphylococcus aureus* and *Candida albicans* [8] and results revealed that thyme has strong inhibitory effect against studied microorganisms [3]. Scientist reported the significant preservative and increased shelf-life effects of wild thyme (*Thymus serpyllum*), about 15 to 20 days on freshwater fish [22]. Scientist reported the antifungal potential of thyme essential oil in complete inhibition of *Fusarium mycelium* [4].
Bread, being the widely consumed product in the world [28], was best chosen product for the supplementation of functional ingredients. For a novel food additive, it was necessary to study its impact on food product quality attributes. Significant influence of food additives on dough properties and product quality had been reported by various researchers [12, 14, 11]. Therefore, the influence of dried thyme on dough and bread quality was investigated. The objectives of this research were to develop thyme enriched functional bread with good shelf life without much affecting the quality characteristics of prepared product.

Material and Methods
Raw materials
Thyme was purchased from local market. It was dried at 50 ºC±1 ºC for 4-5 hour. Dried thyme was packed in sterilized plastic bags and treated in microwave for 915 MHz for 1 min. Wheat flour, yeast, salt, sugar, etc were purchased from local market for bread making.

Physicochemical properties of raw materials
Prescribed protocols of procedures for moisture, ash, protein and fibre were thoroughly followed [1].

Determination of antioxidant capacity
The antioxidant activity of dried thyme was evaluated through free radical scavenging effect on 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical. The determination was based on the standard method [2]. Percentage of DPPH scavenging activity was calculated as% inhibition of DPPH = [(Abs control – Abs sample)/Abs control] × 100.

Determination of total phenolic content
Total phenolic contents of thyme extracts were determined using Folin-Ciocalteu reagent [27]. Samples were inserted into different test tube and mixed thoroughly with 5 mL Folin-Ciocalteu reagent (previously pre-dilute 10 times with distilled water). After 5 min, 4 mL of 7.5% sodium carbonate was added and allowed to react for 2 h at room temperature. The absorbance was measured at 765 nm using spectrophotometer. Samples were measured in three replicates. Standard curve of gallic acid solution (10, 20, 40, 60, 80 and 100 ppm) was prepared using the similar procedure. The results were expressed as mg GAE/100 gm extract sample.

Bread making
Straight dough AACC method was followed for bread making [1]. The formula for control was as: flour-100 gm, compressed yeast-3.0 gm, sugar-2.5 gm, bakery shortening-2.0 gm, salt/NaCl-1.0 gm, potassium bromated 1 ppm, water optimum. The dough was prepared and baking schedule was given as: fermentation-45 min, remixing-25 sec, recovery-20 min, sheeting and moulding-3 min, proofing (@86ºF, RH 75%)-55 min, baking-25 min at 450ºF.

Bread quality
The bread loaves were analyzed for quality attributes. Loaf volume of bread was determined by the seed displacement method [30]. Specific volume was calculated as the ration of the loaf volume to the loaf weight determined an hour after baking according to the standard method [23].

Sensory quality
Sensory evaluation for appearance, color, texture, flavor and overall acceptability was carried out next day by a panel of minimum ten semi trained judges on nine point hedonic scale [20].

Texture analysis
Hardness was measured as an index of bread texture by Stable Micro System Texture Analyzer Model (TA-H di England) using settings as Test-TPA, Probe-75 mm Cylindrical, Pre-test speed-1 mm/s, Test speed-1 mm/s, Post-test speed-1 mm, Force- 50 kg.

Storage studies
Bread samples were packed in low density polyethylene bags and analyzed for visual mold growth for a week at room temperature (30±1 ºC)

Statistical analysis
Data obtained was analyzed statistically using techniques of analysis of variance (ANOVA) [26].

Results and Discussion
Results for chemical composition, total phenolic content (TPC) and antioxidant activity of wheat flour and dried thyme were shown in Table 1. Thyme was found to be rich in TPC (46.50 mg GAE/100g DW) and Antioxidant activity (68.76%) along with appreciable amount of fibre (17.71%).
The farinograph results of thyme supplemented dough and the control (without thyme addition) were shown in Figs. 1 and Table 2. Addition of thyme increased water absorption rate of dough as compared to control. Thyme at 4% level had the highest water absorption (55.60%). High content of fiber (Table 1) increased the water absorption rate of the dough. Scientist also reported increased water absorption with an increase in level of rice bran fiber in dough [24]. There was a decrease in dough stability as thyme level increased in the formulation. Dough stability was reduced due to presence of phenolic acids in thyme. Scientist also reported the similar results [15]. So the baking process had to be modified accordingly. Presence of phenols in thyme had a negative impact on dough stability, mainly at higher levels of incorporation. Results obtained from rheological studied helped to modify the baking procedure to obtain a quality product.

Effect of incorporation of thyme on quality of bread
The baking quality bread was given in Table 3. Bake absorption increased with increase in the level of incorporation of thyme in flour. Thyme had a positive effect on bread quality up to 2% level of supplementation. It was depicted from the higher specific volume values at 1 and 2% (4.86cc/g and 4.79cc/g, respectively) than control (4.72cc/g).
Hardness, namely firmness, was one of the commonly used indices to describe bread quality as the change of hardness normally reflects the loss of resilience [30]. In textural analysis, no significant difference in firmness was observed between bread samples with and without the addition of thyme (Table 3). This indicates that appropriate addition levels of thyme (1-4%) would not cause any undesirable change in hardness of bread. Bread prepared with incorporation of thyme awarded more scores by the panelist up to 2% level of incorporation and showed better overall acceptability (7.98 to 8.06) as compared to control (7.90). Scientist reported an increased acceptability of bread with supplementation with herbs [16]. Results of sensory analysis suggested that addition of thyme in bread formula up to 2% level of incorporation did not interfere in bread acceptability.
Thyme enriched bread (2%) had a total phenolic content (TPC) of 0.67±0.12 GAE/100 g DW and radical scavenging activity (RSA) 15.50±0.09 (Table 4). Similar results were reported by scientist [25]. Data indicated that thyme bread had an increased phenolics amount of approximately three-time compared to the quantity contained in the control. Results of radical scavenging activity were consistent with those obtained from measurement of total phenolics. Scientist reported similar results [18].

Storage study
Breads prepared with best 2% thyme were packed in LDPE and stored at ambient temperature and checked for visible mold growth on daily basis. Effect of incorporation thyme in bread on visual mold growth was given in Table 5. Control bread which contained no preservative spoiled within three days. The thyme bread had a shelf life of 6 days. Inhibitory action of thyme against bakery mold was reported in the literature [21, 13, 19].

Table 1: Chemical composition, total phenolic content (TPC) and antioxidant activity of raw material

Samples	Moisture (%)	Protein (%)	Crude Fibre (%)	TPC (mg GAE/100 g DW)	Antioxidant activity (DPPH% inhibition)
Wheat flour	12.03±0.56	9.17±1.09	0.49±0.08	3.78±0.21	17±2.13
Thyme	11.17±0.12	3.63±0.05	17.71±0.14	46.50±1.09	68.76±1.18

Table 2: Effect of incorporation of dried thyme on farinographic characteristics of flour

Samples	Level (%)	Water absorption (%)	Dough development time (min)	Dough stability (min)	Mixing tolerance index (B.U.)
Control	0	53.00±1.56	1.50±0.05	4.00±0.16	80.0±1.12
Thyme	1	53.70±1.15	2.00±0.04	3.00±0.09	100.0±1.17
	2	53.50±1.25	1.50±0.04	3.00±0.11	125.0±2.50
	3	54.0±2.37	2.00±0.17	2.50±0.22	70.0±1.50
	4	55.60±1.09	2.00±0.15	2.45±0.13	100.0±1.30
CD(p≤0.05)		0.34	0.43	0.48	8.34

Mean ± SD (n = 3)

Table 3: Effect of incorporation of thyme on baking and sensory quality of bread

Samples	Level (%)	Bake absorption (%)	Specific volume (cc/g)	Firmness (kg)	Overall acceptability (score out of 9.0)
Control	0	70.12±1.17	4.72±0.12	1.39±0.05	7.90±0.78
Thyme	1	71.78±1.08	4.86±0.11	1.41±0.06	7.98±1.16
	2	73.57±1.09	4.79±0.05	1.37±0.02	8.06±0.89
	3	74.00±1.15	4.46±0.15	1.32±0.01	7.61±0.45
	4	76.67±1.13	4.24±0.34	1.36±0.05	7.45±0.65
CD(p≤0.05)		0.86	0.18	NS	0.19

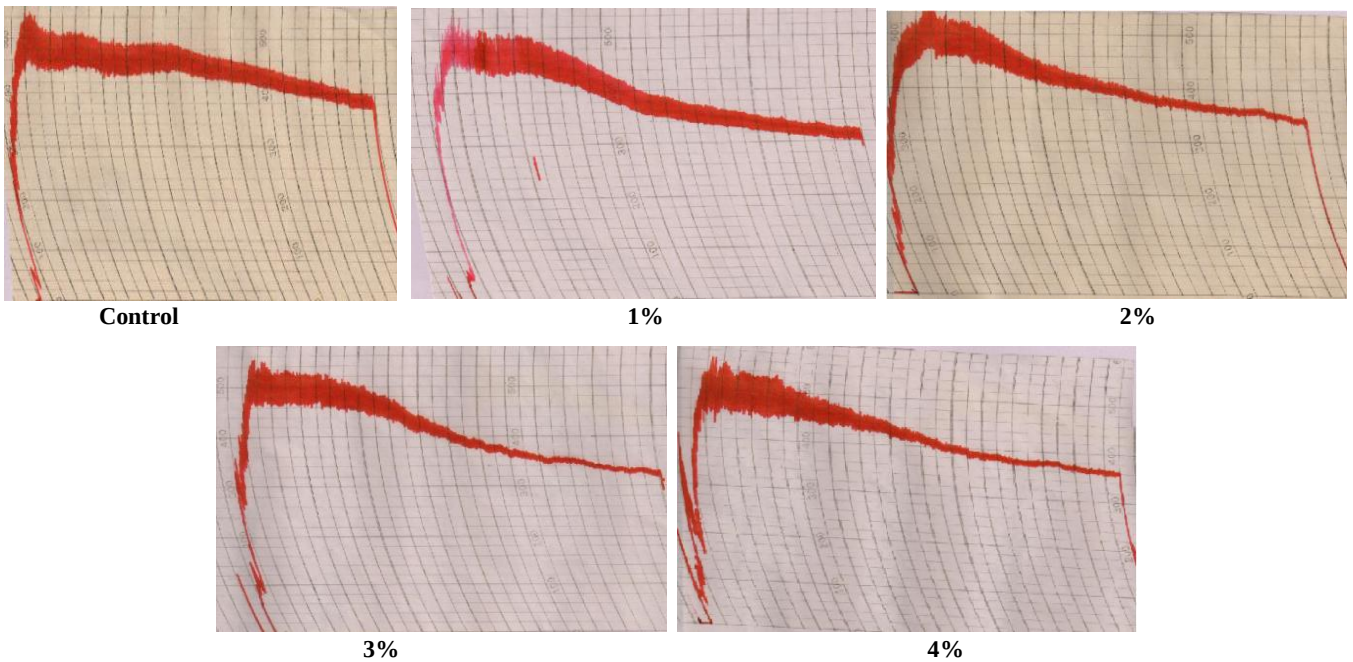
Mean ± SD (n = 3)

Table 4: Effect of incorporation of thyme on total phenolic content and antioxidant activity of bread

Samples	Level (%)	Parameters	
		TPC (mg GAE/100 g DW)	Antioxidant activity (DPPH% Inhibition)
Control	0	0.23±0.02	4.67±0.67
Thyme	1	0.50±0.21	12.15±0.15
	2	0.67±0.12	15.50±0.09
	3	0.75±0.11	17.13±0.55
	4	1.05±0.15	18.34±0.67

Table 5: Effect incorporation of thyme on visual mold growth of bread stored at room temperature (30±1°C)

Samples	Levels (%)	Visual mold growth				
		Days				
		0	2	4	6	8
Control	0	ND*	ND	D**	-	-
Thyme	1	ND	ND	D	-	-
	2	ND	ND	ND	D	-
	3	ND	ND	ND	ND	D
	4	ND	ND	ND	ND	D



Control **1%** **2%**

3% **4%**

Fig 1: Farinograms obtained after addition of dried thyme at different levels.

Conclusions

Thyme addition in wheat flour increased the water absorption and dough development time whereas, dough stability was decreased as level of thyme increased in the blend. Thyme showed a decrease in mixing tolerance index as the levels of thyme increased in flour. An increased baking absorption and specific volume were observed for thyme bread than control. Thyme bread had no significant effect on the texture of bread. Results of sensory analysis suggested that addition of thyme in bread formula up to 2% level of incorporation did not interfere in bread acceptability. Thyme bread was awarded more scores than control and it had a shelf life of 6 days at room temperature (30±1°C). Incorporation of thyme markedly increased the total phenolic content and radical scavenging activity of bread and makes it a functional food.

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