EFFECTS OF CONNECTIVE TISSUE PROTEINS ON CHEMICAL COMPOSITION AND SENSORY PROPERTIES OF GROUND BEEF

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Background

Meat is an important source of protein in the diet and the major meat proteins provide a good balance of the essential amino acids needed by man. However, the connective tissue components of meat, collagen and elastin, have uncharacteristic amino acid compositions, which either lack or present low amounts of several of the essential amino acids. This makes the connective tissue nutritionally poor and could influence the overall quality of meat if they were present in high amounts (BAILEY & LIGHT, 1989). Sensory analysis is an additional method of determining overall quality of meat products by using human tasters in organized and formalized experiments. To characterize sensory and nutritional value of ground meat, it is necessary to know the tissue composition. Besides fat and water content, the major emphasis in quality grading is placed on the muscle protein and connective tissue. The determination of connective tissue is an essential part of every quality control procedure. The term connective tissue covers a wide variety of structures within the body. There are those, such as tendons and ligaments, which clearly connect and hold tissues together, while others, associate more intimately with specific organs. In muscle, for example, connective tissue is present as epi, peri- and endomysial components, which surround and invest the muscle fibers and thus provide the functional integrity, essential for precise muscle action (SIMS and BAILEY, 1981). Connective tissue is a major problem associated with the acceptance of ground beef. The raw ground beef meat regulation allow up to 10% fat and 3% sinew without bone, tendon and cartilages (BRASIL, 1978). With regulation purpose in views, it is necessary to establish the analytical value of collagenous connective tissue proteins that can be present in beef ground meat, in order to ensure their nutritive and sensory value.

Objectives

Evaluate the effects of raw beef connective tissue at 0 to 50% replacement levels on the chemical composition, collagenous connective tissue amount, pH, color values and sensory acceptance of raw ground beef meat. Determine their effects on sensory properties of hamburgers. Offer helpful informations contributing to legislation standards.

Methods

Products manufacture: the fresh beef connective tissue (BCT) consistes largely of epimysium and perymisium removed from bovine muscles obtained from a commercial packing plant that utilizes Skyner machine (Model 7600 - Towsend). The BCT was packaged in vacuum plastic bags and frozen at –18°C. BCT packages were 24h thawed in refrigerator and passed three times through a grinder (CAF 8) plate having 5mm diameter holes. Commercially available biceps femoris muscles were manually trimmed to remove intermuscular fat, epimysial connective tissue, heavy bands of connective tissue within the muscle and grounded through a 5mm plate. BCT was added to ground meat at 3, 5, 10, 15, 20, 25, 30, 35, 40, 45 or 50% levels. The control (0%) contained only grounded lean meat. Every treatment was mixed for 3 min in 2Kg lots using an orbital mixer. The ground meat mixtures were promptly utilized for chemical analysis, pH, objective color measurements and consumer acceptance test. The 10, 20 and 30% BCT formulations were mixed for 1min with 1.0% salt, 0.5% onion soup powder, formed up into 80g hamburgers (10 cm in diameter) and placed in a -18°C freezer until the time of sensory analysis. **Physico-chemical analysis**: moisture, protein (Kjeldahl, Nx6,25), fat (diethyl ether extractable) and ash contents were determined in duplicate following INSTITUTO ADOLFO LUTZ (1985) procedures. pH values were measured with a spear-tip electrode attached to digital pH-meter (HANNA Instruments – HI9321 microprocessor). **Hydroxyproline assay:** was carried out according to the method described by AOAC (1995). Hydroxyproline is quantitatively determined for colagenous material amount measurement. Samples are hydrolyzed with 6N HCl for 8h at 110 degrees C. After hydrolysis, 4-hydroxyproline is converted to pyrrole with chloramine T in acetate-citrate buffer pH 6.0, and pyrrole is converted to a red-coloured complex (absorption at 558nm) by reaction with Ehrlich reagent [p-(dimethylamino)benzaldehyde in perchloric acid/2-propanol]. **Total connective tissue proteins:** were determined multiplying the hydroxyproline contents by 8. **Color:** five objective color readings were taken from ground meat mixture surfaces using a spectrophotometer Minolta CM-508d, with Illuminant D_{65} and 2° standard observer. The results were expressed as CIE-Lab L* (lightness), a* (redness), b* (yellowness). **Sensory evaluation**: a trained 11-member panel rated raw ground meat mixtures formulated with 10, 20 and 30% BCT for red color and sinew amount, using a unstructured 10 cm line. The attributes firmness, amount of residual connective tissue remaining at the end of the chewing process, characteristic flavor and masticability (chews number) were evaluated in hamburgers roasted in a 260-280°C electric oven for 20 min, quartered, and served as hot as possible to the panelists in booths under red lights with controlled temperature. Replicate judgments were made among all assistants. **Consumer acceptance test:** a consumer panel test was carried out for 57 panelists. Five numbered ground meat mixture 0, 10, 20, 30 and 40% BCT replacement were presented in booths with controlled temperature. The appearance acceptability was evaluated on a 9 points verbal hedonic scale, sinew amount in a 9 points just right scale and purchase intent in a 5 points scale. **Statistical analysis**: the sensory design was randomized complete block. Results were statistically analyzed by ANOVA and Tukey test of 5% level of significance using GraphPad InStat Software v2.01 (1990-1993).

Results and Discussion

Based on Table 1 results, for raw beef connective tissue (BCT) at 0 to 50% replacement levels to ground beef meat, ranges of percent moisture were 76.46-70.04%, fat 2.24-8.40%, total protein 19.84-20.58%, ash 1.03-0.91%, hydroxyproline 0.08-0.52%, collagenous connective tissue 0.62-4.15%, collagenous connective tissue per total protein 3.12-20.16%, pH 5.81-5.65, L* value 36.02-45.14, a* value 19.42-14.71 and b* value 4.68-9.85. The addition of 20% BCT decreased moisture, increased fat and L* (lightness) when compared to the control 0% ($p<0.05$). BCT added at 15% level increased ($p<0.05$) 4-Hydroxyproline and collagenous connective tissue compared to the control. Addition of BCT had a slight increase effect on total protein of ground meat ($p>0.05$). Ash was affected with 30% BCT replacement. BCT had a significant effect on pH, but the actual range in pH value (0.16 units) was of little practical importance. Color a* (redness) and b*(yellowness) values were significantly different from control at 35% BCT level. As assessed by sensory panel (Table 2), 20% BCT added to ground meat significantly ($p<0.05$) reduced red color intensity, increased sinew amount (appearance) and hamburger firmness. Hamburgers added with 30% BCT showed increased masticability, connective tissue residue remaining at the end of mastication and decreased characteristic flavor. Laboratory consumer test (Table 3) revealed that 20% BCT in ground meat resulted in a slightly disliked appearance; moderately too much sinew amount and this product maybe/maybe not would buy. Figure 1 shows closer linear relationship between collagenous connective tissue (R2=0,985) or collagenous connective tissue per total protein (R2= 0,973) and predicted beef connective tissue.

Conclusions

In general, fresh beef connective tissue (BCT) added to beef ground meat decreases values and contents of moisture, ash, pH, a* (redness), increases fat, hydroxyproline, collagenous connective tissue, collagenous connective tissue per total protein, L* (lightness) and b*(yellowness). There was a definite linear relationship between the amount of connective tissue added and determined. These findings suggest that analytical collagenous connective tissue content can be a useful parameter for regulatory evaluation of connective tissue replacement in beef ground meat. Connective tissue contributed to increase hamburger firmness, masticability, as well as to decrease red color and characteristic flavor. Addition of 10% BCT ground meat resulted in acceptable product appearance; more connective tissue replacement produced less acceptable ground meat.

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Table 1. Chemical composition and physical parameters means for ground meat containing beef connective tissue

Parameters	Beef connective tissue replacement in ground meat (%)												
	0	3	5	10	15	20	25	30	35	40	45	50	100
Moisture (%)	76.46^{a}	75.82^{a}	75.31$^{a.b}$	75.39$^{a.b}$	74.92$^{a.b}$	73.54$^{b.c}$	73.67$^{b.c}$	72.28$^{c.d}$	71.65$^{c.d.e}$	71.00$^{d.e}$	70.70$^{d.e}$	70.04^{e}	62.92^{f}
	(0.12)	(0.82)	(0.33)	(0.17)	(0.42)	(0.87)	(0.75)	(0.02)	(0.23)	(0.36)	(0.77)	(0.06)	(0.71)
Fat (%)	2.24^{g}	2.58$^{f.g}$	2.92$^{f.g}$	4.30$^{e.f.g}$	3.96$^{e.f.g}$	4.70$^{d.e.f}$	5.63$^{c.d.e}$	6.67$^{b.c.d}$	6.70$^{b.c.d}$	7.72$^{b.c}$	8.33^{b}	8.40^{b}	15.45^{a}
	(0.11)	(0.12)	(0.39)	(0.31)	(0.06)	(0.02)	(1.11)	(0.23)	(0.45)	(0.43)	(0.88)	(1.19)	(0.49)
Total protein (%)	19.84^{b}	19.88^{b}	20.01^{b}	19.96^{b}	20.04^{b}	20.02^{b}	19.48^{b}	19.96^{b}	19.85^{b}	20.02^{b}	20.26$^{a.b}$	20.58$^{a.b}$	21.76^{a}
	(0.42)	(0.31)	(0.42)	(0.34)	(0.65)	(0.16)	(0.06)	(0.31)	(0.47)	(0.54)	(0.61)	(0.08)	(0.32)
Ash (%)	1.03^{a}	1.00$^{a.b}$	1.00$^{a.b.c}$	1.02^{a}	1.02^{a}	1.00$^{a.b}$	0.99$^{a.b.c}$	0.94$^{b.c.d}$	0.94$^{b.c.d}$	0.94$^{b.c.d}$	0.93$^{c.d}$	0.91^{d}	0.77^{e}
	(0.01)	(0.04)	(0.02)	(0.00)	(0.01)	(0.01)	(0.03)	(0.01)	(0.00)	(0.01)	(0.01)	(0.02)	(0.02)
Hydroxyp. (%)	0.08^{f}	0.10$^{e.f}$	0.13$^{e.f}$	0.17$^{e.f}$	0.22$^{d.e}$	0.32$^{c.d}$	0.36^{c}	0.43$^{b.c}$	0.44$^{b.c}$	0.51^{b}	0.50^{b}	0.52^{b}	1.01^{a}
	(0.03)	(0.00)	(0.02)	(0.00)	(0.02)	(0.09)	(0.04)	(0.01)	(0.03)	(0.03)	(0.01)	(0.01)	(0.03)
Collagenous con. tissue (%)	0.62^{f}	0.79$^{e.f}$	1.06$^{e.f}$	1.36$^{e.f}$	1.74$^{d.e}$	2.55$^{c.d}$	2.87^{c}	3.42$^{b.c}$	3.50$^{b.c}$	4.09^{b}	4.02^{b}	4.15^{b}	8.09^{a}
	(0.26)	(0.03)	(0.18)	(0.01)	(0.128)	(0.71)	(0.29)	(0.06)	(0.26)	(0.25)	(0.10)	(0.13)	(0.21)
Coll. con.tis. per tot. protein (%)	3.12	3.97	5.30	6.81	8.68	12.74	14.73	17.13	17.63	20.43	19.84	20.16	37.18
pH value	5.81^{a}	5.80$^{a.b}$	5.76$^{b.c}$	5.76$^{b.c}$	5.76$^{b.c}$	5.74$^{c.d}$	5.75$^{b.c}$	5.68$^{d.e.f}$	5.67$^{e.f}$	5.65^{f}	5.67$^{e.f}$	5.65^{f}	5.71$^{c.d.e}$
	(0.01)	(0.01)	(0.01)	(0.01)	(0.01)	(0.01)	(0.01)	(0.00)	(0.01)	(0.03)	(0.01)	(0.01)	(0.01)
L* value	36.02^{f}	36.50$^{e.f}$	37.45$^{d.e.f}$	38.06$^{d.e.f}$	39.54$^{c.d.e.f}$	42.17$^{b.c.d.e}$	42.16$^{b.c.d.e}$	43.01$^{b.c.d}$	44.90$^{b.c}$	47.01^{b}	45.82^{b}	45.14$^{b.c}$	54.71^{a}
	(1.02)	(1.77)	(2.84)	(2.11)	(2.70)	(2.76)	(1.83)	(4.56)	(3.50)	(1.96)	(3.71)	(3.13)	(1.23)
a* value	19.09$^{a.b}$	17.09$^{a.b.c}$	19.42^{a}	18.52$^{a.b.c}$	17.89$^{a.b.c}$	17.55$^{a.b.c}$	16.50$^{a.b.c}$	16.29$^{a.b.c}$	15.35$^{b.c.d}$	14.71$^{c.d}$	15.71$^{a.b.c}$	15.69$^{a.b.c}$	11.45^{d}
	(0.28)	(1.22)	(1.10)	(2.20)	(1.47)	(1.00)	(1.80)	(2.19)	(3.11)	(1.86)	(1.59)	(2.67)	(1.23)
b* value	5.67^{d}	4.68^{d}	6.48$^{c.d}$	5.72$^{c.d}$	6.75$^{b.c.d}$	7.58$^{b.c.d}$	7.75$^{b.c.d}$	7.71$^{b.c.d}$	8.89$^{a.b.c}$	9.21$^{a.b}$	9.85$^{a.b}$	9.51$^{a.b}$	10.99^{a}
	(0.81)	(0.84)	(2.62)	(0.61)	(0.98)	(1.19)	(1.86)	(2.47)	(1.31)	(0.66)	(1.75)	(1.14)	(0.60)

( )Standard deviation ^{a,b,c,d,e,f} Means on the same horizontal row with different superscripts are significantly different (p<0,05)

Table 2. Mean sensory scores for beef hamburgers

Sensory attributes*	Beef connective tissue (BCT) replacement in hamburgers (%)		
	10	20	30
Red color[1]	6,38^{a} (0,28)	4,31^{b} (0,36)	3,01^{c} (0,40)
Sinew amount[2]	4,15^{c} (0,40)	6,35^{b} (0,23)	7,90^{a} (0,26)
Firmness[3]	5,31^{b} (0,35)	6,42^{a} (0,32)	7,04^{a} (0,27)
Connective tissue amount[4]	4,76^{b} (0,48)	5,58^{b} (0,47)	6,66^{a} (0,32)
Characteristic flavour[5]	6,88^{a} (0,47)	5,93$^{a.b}$ (0,49)	5,52^{b} (0,46)
Mastigability[6]	36^{b} (3)	37^{b} (4)	44^{a} (5)

* Means of 11 judgments using 10cm linear scale: [1]0=slight; 10=intense; [2]0=none; 10=very abundant; [3]0=not firm; 10=very firm; [4]0=none; 10=abundant; [5]0=slight; 10=intense; [6]Number of chews

^{a,b,c}Means on the same horizontal row with different superscripts are significantly different (p<0,05) ( ) Standard error of the mean

Table 3. Laboratory consumer test for ground beef meat added with beef connective tissue (BCT)

Sensory attributes	Beef connective tissue (BCT) replacement in ground meat (%)				
	0	10	20	30	40
Hedonic scale[1]	7,85^{a}	6,24^{b}	4,29^{c}	2,56^{d}	1,47^{e}
Appearance acceptability	(0,16)	(0,21)	(0,20)	(0,14)	(0,08)
Just right scale[2]	-0,21^{e}	1,02^{d}	2,21^{c}	2,87^{b}	3,74^{a}
Sinew amount	(0,09)	(0,13)	(0,12)	(0,13)	(0,08)
Purchase intent scale[3]	4,63^{a}	3,93^{b}	2,56^{c}	1,75^{d}	1,10^{e}
	(0,12)	(0,10)	(0,12)	(0,11)	(0,04)

[1]9=like extremely; 5=neither like not dislike; 1= dislike extremely

[2]+4=extremely too much; 3=much too much; 2= moderately too much; 1= somewhat too much; 0= just right; -1= somewhat too little; -4=extremely too little

[3] 5=definitely would buy; 4=probably would buy; 3=maybe/maybe not, 2=probably would not buy; 1=definitely would not buy ^{a,b,c}Means on the same horizontal row with different superscripts are significantly different (p<0,05) - 57 judgments

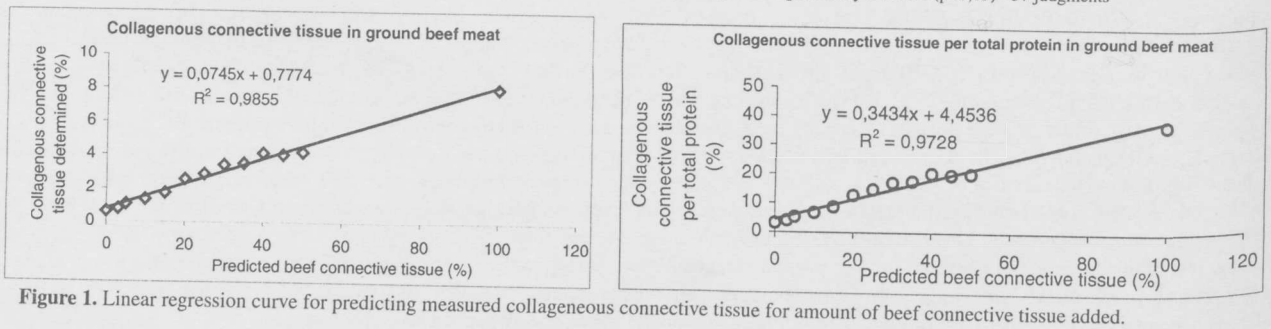

Figure 1. Linear regression curve for predicting measured collageneous connective tissue for amount of beef connective tissue added.