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Comparative evaluation of the amino acid profile of the muscle and skin of guinea fowl (*Numida meleagris)* hen

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ABSTRACT

The amino acid composition of the muscle and skin of the matured female guinea fowl (*Numida meleagris*) was determined on a dry weight basis. The total essential amino acids ranged from 30.4 g/100g to 43.5 g/100g crude protein or from 49.7% - 51.2% of the total amino acid. The amino acid score showed that lysine ranged from 0.66-1.17 (on whole hen's egg comparison), 0.75-1.31 (on provisional essential amino acid scoring pattern) and 0.71-1.25 (on suggested requirement of the essential amino acid of a preschool child). The predicted protein efficiency ratio was 1.81-2.25 and the essential amino acid index range was 0.87-1.28. The correlation coefficient (r_{xy}) was positive and significant at $r_{=0.05}$ for the total amino acids, amino acid scores (on whole hen's egg basis) and other parameters in the two samples. Results have good comparison with whole hen's egg protein.

Introduction

For Third World villages, the guinea fowl (*Numida meleagris)* could become much more valuable than it is today. The bird thrives under semi-intensive conditions, forages well, and requires little attention. It retains many of its wild ancestor's survival characteristics: it grows, reproduces, and yields well in both cool and hot conditions; it is relatively diseased free; it requires little water; it is almost as easily raised as chickens and turkeys; and it is a most useful all-round farm bird[1].

The guinea fowl's potential to increase meat production among hungry countries should be given greater recognition. The birds are widely known in Africa and occur in a few areas of Asia, but they show promise for use throughout all of Asia and Latin America and for increased use in Africa itself. Strains newly created for egg and meat production in Europe – notably in France – show excellent characteristics for industrial – scale production. Also, many semi domestic types in Africa deserve increased scientific assessment as scavenger birds.

Meat from domestic guinea fowl is dark and delicate, the flavour resembling that of game birds. It is a special delicacy, served in some of world's finest restaurants. Several European countries eat vast amounts. Annual consumption in France, for example is about 0.8kg per capita[1].

Guinea fowl also produce substantial numbers of eggs. In Africa, these are often sold hard-boiled in local markets. In the Soviet Union, they are produced in large commercial operations. In France, guinea fowl strains have been developed that not only grow quickly but lay as many as 190 eggs a year.

Outside Europe, virtually all guinea fowl are raised as free-ranging birds. These find most of their feed by scratching around villages and farmyards. Their cost of production is small, and they yield food for subsistence farmers. In Europe, in the main, guinea fowl is raised to produce meat for luxury market.

Guinea fowl production is beginning to increase all over the world. There are no reports on the chemical composition of female guinea fowl meat. Due to the emphasis placed on the nutritive value of food by consumers a great need exists for information on nutritional composition of guinea fowl meat. The present study was therefore undertaken in attempt to gain some information on the amino acid of the muscle and skin. The skin was being analysed separately to know its nutritive value because it is often suggested to be discarded in order to avoid the fat from guinea fowl meat. The guinea fowl sample used was the pearl type.

Materials and methods

The guinea fowl hen used was a matured bird. Prior to butchering, food was withheld for a day to help ensure the digestive system was empty. Head was held on the stump and the guinea head removed with an axe. At the end of bleeding, the guinea was plucked. When all the feathers were removed, the guinea fowl's anus was rinsed to remove any residue, and then a sharp knife was inserted just below the hip bone without puncturing any of the internal organs. The guinea was then removed; both skin and muscle sliced, rinsed and dried in the oven. Muscle and skin used were those from the breast part. The dried samples were ground, sieved and kept in freezer in McCartney bottles pending analysis.

The micro-Kjeldahl method as described by Pearson[2] was followed to determine the fat-free crude protein. The fat was extracted with a chloroform/methanol (2:1) mixture using Soxhlet extraction apparatus[3]. Between 30mg and 35mg deffated samples were weighed into the glass ampoule. Then 7 ml of 6 M HCl was added and oxygen was expelled by passing nitrogen gas into the ampoule. The glass ampoules were sealed with flame and put in an oven preset at 105±5^{o}C for 22 h. The ampoules were cooled and opened and the contents were filtered. The filtrate was evaporated to dryness at 40^{o}C under vacuum. The residue was dissolved with 5 ml acetate buffer (pH 2.0) and stored in the freezer.

Amino acid analysis was by ion-exchange chromatography[4] using a Technicon Sequential Multisample Amino Acid Analyzer (Technicon Instruments Corporation, New York,

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USA). The period of analysis was 76 min, with a gas flow rate of 0.50 ml/min at 60°C, and the reproducibility was ±3%. The amino acid values were the average of two determinations. Tryptophan was not determined. Amino acid scores were calculated using three different procedures: (i) scores based on amino acid values compared with whole hen's egg amino acid profile[5]; (ii) scores based on essential amino acid scoring pattern[6]; (iii) scores based on essential amino acid suggested pattern of requirements for preschool child[7]. The predicted protein efficiency ratio (P-PER)[8], using an equation of the form P-PER = -0.468 + 0.454 (Leu) – 0.105 (Tyr). The theoretical estimation of isoelectric point (pI) was determined using the equation[9,10]:

$$IPm = \sum_{i=1}^{n} PiXi$$

where IPm is the isoelectric point of the i^{th} amino acid in the mixture and Xi is the mass or mole fraction of the i^{th} amino acid in the mixture. The essential amino acid index (EAAI) was determined using the method of Steinke et al.[11]. The leucine/isoleucine ratios, their differences and their percentage differences were also calculated.

The statistical analysis carried out included the determination of the grand mean, standard deviation (SD) and the coefficients of variation percent (CV %). Other calculations made were the simple linear correlation coefficient (r_{xy}), coefficient of determination (r_{xy}^2), coefficient of alienation (or index of lack of relationship) (C_A) and index of forecasting efficiency (IFE) and to test for significance difference, the level of probability was set at $r_{=0.05}$ at n-2 degrees of freedom[12].

Results and discussion

Table I presents the amino acid composition of the samples. Glu was the most concentrated amino acid (AA) in both muscle and skin with values of 13.3g/100g crude protein (cp) in the muscle and 8.60g/100g cp in the skin. A look at Table I will show that AA of the muscle is more concentrated (on pairwise comparison) than the corresponding AA in the skin. The Asp was the second largest AA in the two samples. The most concentrated essential AA (EAA) in the samples was Lys (7.23g/100gcp) in the muscle and Arg (5.26g/100g cp) in the skin. The coefficient of variation percent (CV %) ranged between 4.33 to 53.8 in the AA, with the Tyr having the least CV% and Ser the highest CV%. Sales and Hayes[13] studied the proximate, amino acid and mineral composition of ostrich meat; the meat was the ostrich muscles *iliofibularis, femorotibialis medius* and *gastrocnemius pars interna*. Ostrich meat when compared with the guinea hen muscle had the following results (present study/literature value) in g/100gcp: Lys (7.23/8.3 – 8.72), His (2.51/1.95-2.07), Arg (7.00/6.77-7.27), Asp (9.96/9.66-9.99), Thr (3.40/3.82-3.96), Ser (4.0/3.00-3.06), Glu (13.3/15.5-16.1), Gly (5.10/4.02-4.05), Ala (4.20/5.28-5.65), Met (3.50/2.74-2.93), Val (5.52/4.75-5.35), Ile (3.39/4.58-4.96), Leu (6.72/8.43-9.11), Phe (4.20/4.71-5.07) and Tyr (3.20/3.09-3.17).Sales and Hayes[13] did not report on Cys, Pro and Try; among those AA reported, the levels of His, Ser, Gly, Met, Val and Tyr were higher in the muscle of guinea fowl than the muscle of ostrich while the rest AA are favourably comparable. On the other hand, the guinea fowl muscle was better concentrated in Val, Met, Arg and Asp AA than in beef species[14] whereas Gly, Val, Met, Arg, Asp and Ser levels in guinea fowl meat were higher than their corresponding levels in chicken[14]. On comparison of the muscle of guinea fowl hen with the muscle of turkey-hen, the following AA were better concentrated in the guinea: Lys, Gly, Ala, Cys, Val, Met and Leu whereas Pro had similar level with turkey hen muscle; also while the fat-free dry weight protein was 84.2g/100g in turkey hen; it is 84.4g/100g in guinea fowl muscle; the skin of guinea fowl was better concentrated in Tyr and Ala than in the turkey skin[15]. The Cys level in beef was 1.1g/100g cp[16] which is lower than the value in guinea fowl muscle. The guinea fowl muscle was better in Met, Phe and Val concentration than in pork; better in Met, Cys, Phe and Val than in mutton[16]. With the exception of Leu, the pattern of AA concentration in the guinea fowl hen muscle and skin followed the trend observed in the muscle and skin of the turkey hen[15].

The FAO/WHO/UNU[7] standards for pre-school children (2-5 years) were (g/100g protein): Leu (6.6), Phe + Tyr (6.3), Thr (3.4), Try (1.1), Val (3.5), Ile (2.8), Lys (5.8), Met + Cys (2.5), His (1.9) and total (33.9 with His) and 32.0 (no His). Based on this information, the muscle would provide enough or even more than enough of His, Ile, Leu, Met + Cys, Phe + Try, Thr, Val and Lys while skin would provide enough or even more of Ile, Met + Cys and Phe + Tyr. Tryptophan was not determined.

Table II presents parameters on the quality of the protein of the samples. The EAA ranged between 43.5-30.4g/100g cp with a variation of 25.1%. These values were close to the values of 56.6g/100g cp of the egg reference protein[5]. The total sulphur amino acids (TSAA) of the samples were 4.74 g/100g cp (muscle) and 2.91g/100g cp (skin). The value of 4.74g/100g cp was close to the value of 5.8g/100g cp while 2.91g/100g cp formed about one-half recommended for infants[7]. The aromatic AA (ArAA) range suggested for ideal infant protein (6.8-11.8g/100g cp)[7] was very favourably comparable with the current report of 6.61-7.40g/100g cp showing that the samples protein could be used to supplement sorghum flour. The percentage ratio of EAA to the total AA (TAA) in the samples ranged between 49.7% and 51.2%. These values were well above the 39% considered adequate for ideal protein food for infants, 26% for children and 11% for adults[7]. The percentage of EAA/TAA for the samples could be favourably compared with other animal protein sources – 46.2% in *Zonocerus variegatus*[17], 43.7% in *Macrotermes bellicosus*[18] and 54.8% in *Gymnarchus niloticus* (Trunk fish)[19] whereas it is 50% for egg[20]. The TEAA in these results were close to the value of 44.4 g/100g cp in soya bean[21]. The percentage of total neutral AA (TNAA) ranged from 54.3-56.9, indicating that these formed the bulk of the AA; total acid AA (TAAA) ranged from 24.7 – 26.6 which was lower than % TNAA, while the percentage range in total basic AA (TBAA) was 18.5-19.1 which made them the third largest group among the samples.

The predicted protein efficiency ratio (P-PER) was 2.25 (muscle) and 1.81 (skin). These results were exactly opposite the results observed in turkey hen with 2.27 (skin) and 1.93 (muscle)[15]; it is 2.22 (*Clarias anguillaris*), 1.92 (*Oreochromis niloticus*) and 1.89 (*Cynoglossus senegalensis*)[22] but lower than in the values from various parts of fresh water female crab: 3.4 (whole body), 3.1 (flesh), 2.6 (exoskeleton)[23]; fresh water male crab: 2.9 (whole body), 2.8 (flesh), 2.4 (exoskeleton)[24]; 4.06 (corn *ogi*) and reference casein with PER of 2.50[25]. Other literature values were 1.21 (cowpea), 1.82 (pigeon pea)[26]; 1.62 (millet *ogi*) and 0.27 (sorghum *ogi*)[25]. The Leu/Ile ratio was low in both samples (1.91-1.98), this is much less than in turkey hen

(2.65-3.33)[15], hence no concentration antagonism might be experienced in the guinea fowl hen meat. The essential AA index (EAAI) ranged from 0.87 (skin) – 1.28 (muscle). EAAI is useful as a rapid tool to evaluate food formulations for protein quality, although it does not account for differences in protein quality due to various processing methods or certain chemical reactions[27]. The EAAI of defatted soya flour is 1.26[27]; this is close to the muscle EAAI of guinea. In the results of the isoelectric points (pI), there was a shift from 3.47 in skin to 5.11 in the muscle. This type of shift was also observed in turkey meat: from 4.41 in skin to 5.01 in the muscle[15]. The calculation of pI from amino acids would assist in the production of the protein isolate of an organic product.

Most animal proteins are low in Cys, for examples (Cys/TSAA)%: 36.3% in *M. bellicosus*[18]; 25.6% in *Z.variegatus*[17]; 35.5% in *Archachatina marginata,* 38.8% in *Archatina archatina* and 21.0% in *Limicolaria* sp. respectively[28]; 27.3%-32.8% in female fresh water crab body parts[23]; 23.8%-30.1% in three different Nigeria fishes[22]; 13.3% - 15.9% in male fresh water crab body parts[24]; 26.0% - 26.5% in turkey hen meat[15]. The present results corroborated these literature observations with values of 26.2-30.3%. In contrast, many vegetable proteins contain substantially more Cys than Met, examples: 62.9% in coconut endosperm[29] and in *Anacardium occidentale* it is 50.5%[30]. Thus for animal protein diets, or mixed diets containing animal protein, Cys is unlikely to contribute up to 50% of the TSAA[4]. The percentage of Cys in TSAA had been set at 50% in rat, chick and pig diets[4]. Cys has positive effects on mineral absorption particularly zinc[31, 32].

Table III shows the AA scores (AAS) of the samples based on whole hen's egg profile[5]. The scores had values greater than 1.0 in Lys, His, Arg, Glu, Gly, Ala and Met in the muscle but no AA could measure up to this level in the skin. Gly had the highest score (1.70) in the muscle as well as in the skin (0.87); the least score was Ser (0.51) in muscle and also Ser (0.23) in the skin. The guinea fowl meat showed very good comparison with the AA profile of the whole hen's egg. The CV% between AA levels of muscle and skin ranged between 4.53-53.5. Table IV shows the essential AA scores (EAAS) based on the provisional essential amino acid scoring pattern[6]. EAAS greater than 1.0 in muscle were Lys, Met + Cys, Val and Phe + Tyr; it was only Phe + Tyr in the skin. The limiting AA (LAA) in the muscle was Ile (0.848) whereas it was Thr (0.51) in the skin. To make corrections for the LAA in the samples if they serve as sole sources of protein food, it would be 100/84.8 x protein of muscle and 100/51 x protein of skin; or 1.18 x protein of muscle and 1.96 x protein of skin. The highest EAAS in muscle was Met + Cys (1.35) and Phe + Tyr (1.10) in the skin. The Table V shows the EAAS based on suggested requirement of the EAA of a preschool child[7]. It is very interesting to note that all the EAAS in the muscle were greater than 1.0 or equal to 1.0 and Met + Cys, Ile and Phe + Tyr were in that position in the skin. As in Table IV Met + Cys had the highest EAAS (1.90) in the muscle, but unlike in Table IV, Met + Cys (1.16) also occupied the same position in the skin. The LAA in the muscle was Thr (1.00) but needs no special correction; whereas in the skin Thr as the LAA (0.60) would need a correction of 100/60 x protein of skin or 1.67 x protein of skin.

The following values would show the position of the quality of the guinea hen muscle and skin protein; the EAA requirements across board are (values with His) (g/100g protein): infant (46.0), pre-school (2-5 years) (33.9), school child (10-12 years) (24.1) and adult (12.7) and without His: infant (43.4), pre-school (32.0), school child (22.2) and adult (11.1)[7]; from the present results based on these standards, we have: 44.4g/100g protein (with His) and 41.9 (no His) in muscle; 28.5g/100g (with His) and 26.9 (no His) in skin. The muscle results were close to the levels of the total EAA in egg: 51.2 (with His) and 47.7 (no His); beef: 47.9 (with His) and 44.5 (no His)[7] and better than the muscle of turkey hen: 35.5g/100g (with His)and 32.9 (no His) but the turkey hen was better in skin with values of 35.4g/100g (with His) and 32.9 (no His)[15].

Table VI gave a brief summary of the amino acid profile in the two samples. Column under Factor B means show that the values there were very close with a range of 36.6-37.0. However, Table VII depicts the summary of the statistical analysis of results in Tables I, II(pI), III, IV, and V. The simple linear correlation coefficient (r_{xy}) values showed high positive and significant results from Tables I, II and III at $r_{=0.05}$ and n-2 degrees of freedom. Results from Tables IV and V had positive but insignificant r_{xy}.

The regression coefficient (R_{xy}) showed that for every unit increase in the muscle AA parameter, the increase in skin was 1.36 (Table I), 1.35 (Table II, pI), 0.20 (Table III), 0.24 (Table IV) and 0.39 (Table V). The coefficient of alienation (C_A) was low in Table I (0.10 or 10%), Table II (0.35 or 35%) but high in Tables III (0.89 or 89%), IV (0.84 or 84%) and V (0.81 or 81%). The index of forecasting efficiency (IFE) was high in Tables I (0.90 or 90%) and II (0.65 or 65%) while others were low at between 11-19% (Tables III-V). Low IFE versus high C_A makes prediction of relationship difficult. The C_A produces an index of lack of relationship while the IFE gives the reduction in errors of prediction or relationship. The C_A and IFE values showed that a good relationship existed between the muscle and skin in *Numida meleagris* particularly with the results in Tables I, II and III. The pattern of r_{xy} results from Tables I and II were similar to those obtained for the amino acid profiles of the shell and flesh of *Penaeus notabilis*[33].

Conclusion

Numida meleagris hen meat (muscle and skin) was found to be a good source of high quality protein of almost adequate or more than adequate of EAA, high P-PER and low Leu/Ile ratios particularly the muscle thereby providing premium quality meat.

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Table I. Amino acid composition (g/100g crude protein) of guinea hen (dry weight)

Amino acid	Muscle	Skin	Mean	SD	CV%
Lys[a]	7.23	4.11	5.67	2.21	38.9
His[a]	2.51	1.60	2.06	0.64	31.3
Arg[a]	7.00	5.26	6.13	1.23	20.1
Asp	9.96	6.05	8.01	2.76	34.5
Thr[a]	3.40	2.04	2.72	0.96	35.4
Ser	4.01	1.80	2.91	1.56	53.8
Glu	13.3	8.60	11.0	3.32	30.4
Pro	3.05	2.13	2.59	0.65	25.1
Gly	5.10	2.60	3.85	1.77	45.9
Ala	4.20	4.00	4.10	0.14	3.45
Met[a]	3.50	2.03	2.77	1.04	37.6
Cys	1.24	0.88	1.06	0.25	24.0
Val[a]	5.52	3.00	4.26	1.78	41.8
Ile[a]	3.39	3.00	3.20	0.28	8.63
Leu[a]	6.72	5.24	5.98	1.05	17.5
Phe[a]	4.20	3.60	3.90	0.42	10.9
Tyr	3.20	3.01	3.11	0.13	4.33
Try[a]	-	-	-	-	-
Protein (fat free)	84.4	35.1	59.8	34.9	58.3

[a]Essential amino acid. -, not determined.

Table II. EAA, non-EAA, acidic, neutral, sulphur and aromatic acid contents (g/100g crude protein) of guinea hen (dry weight).

Amino acid	Muscle	Skin	Mean	SD	CV%
Total amino acids	87.5	59.4	73.5	19.9	27.1
Total non- essential amino acid(TN EAA)	44.1	29.1	36.6	10.6	29.0
Total EAA					
With His	43.5	30.4	37.0	9.26	25.1
No His	41.0	28.8	34.9	8.63	24.7
% TN EAA	50.4	49.0	49.7	0.99	1.99
% Total EAA					
With His	49.7	51.2	50.5	1.06	2.10
No His	46.9	48.5	47.7	1.13	2.37
Total neutral amino acid (TNAA)	47.5	33.8	40.7	9.69	23.8
% TNAA	54.3	56.9	55.6	1.84	3.31
TAAA	23.3	14.7	19.0	6.08	32.0
% TAAA	26.6	24.7	25.7	1.34	5.24
Total basic amino acid (TBAA)	16.7	11.0	13.9	4.03	29.1
% TBAA	19.1	18.5	18.8	0.42	2.26
Total sulphur amino acid (TSAA)	4.74	2.91	3.83	1.29	33.8
% TSAA	5.42	4.90	5.16	0.37	7.13
% Cys in TSAA	26.2	30.2	28.2	2.83	10.0
Total aromatic amino acid (TArAA)	7.40	6.61	7.01	0.56	7.97
% TArAA	8.46	11.1	9.78	1.87	19.1
P-PER	2.25	1.81	2.03	0.31	15.3
Leu/Ile ratio	1.98	1.91	1.95	0.05	2.54
Leu-Ile (difference)	3.33	2.24	2.79	0.77	27.7
% Leu-Ile difference)	49.6	47.6	48.6	1.41	2.91
EAAI	1.28	0.87	1.08	0.29	27.0
Isoelectric point(pI)	5.11	3.47	4.29	1.16	27.0

Table III Amino acid scores of the guinea hen based on whole hen's amino acid profile

Amino acid	Muscle	Skin	Mean	SD	CV%
Lys	1.17	0.66	0.92	0.36	39.4
His	1.05	0.67	0.86	0.27	31.2
Arg	1.15	0.86	1.01	0.21	20.4
Asp	0.93	0.57	0.75	0.25	33.9
Thr	0.67	0.40	0.54	0.19	35.7
Ser	0.51	0.23	0.37	0.20	53.5
Glu	1.11	0.72	0.92	0.28	30.1
Pro	0.80	0.56	0.68	0.17	25.0
Gly	1.70	0.87	1.29	0.59	45.5
Ala	1.05	0.74	0.90	0.22	24.5
Cys	0.69	0.49	0.59	0.14	24.0
Val	0.74	0.40	0.57	0.24	42.2
Met	1.09	0.63	0.86	0.33	37.8
Ile	0.61	0.54	0.58	0.05	8.61
Leu	0.81	0.69	0.75	0.08	11.3
Tyr	0.80	0.75	0.78	0.04	4.53
Phe	0.82	0.71	0.77	0.08	10.2

Table IV. Amino acid scores of the guinea hen based on the provisional essential amino acid scoring pattern.

Amino acid	Muscle	Skin	Mean	SD	CV%
Lys	1.31	0.75	1.03	0.40	38.4
Thr	0.85	0.51	0.68	0.24	35.4
Met + Cys	1.35	0.83	1.09	0.37	33.7
Val	1.10	0.60	0.85	0.35	41.6
Ile	0.848	0.75	0.80	0.07	8.67
Leu	0.96	0.749	0.85	0.15	17.5
Phe + Tyr	1.23	1.10	1.17	0.09	7.89
Try	-	-	-	-	-
Total	1.17	0.81	0.99	0.25	25.7

Table V. Amino acid scores of the guinea hen based on the suggested requirement of the essential amino acid of a preschool child.

Amino acid	Muscle	Skin	Mean	SD	CV%
Lys	1.25	0.71	0.98	0.38	39.0
His	1.32	0.84	1.08	0.34	31.4
Thr	1.00	0.60	0.80	0.28	35.4
Val	1.58	0.86	1.22	0.51	41.7
Met + Cys	1.90	1.16	1.53	0.52	34.2
Ile	1.21	1.07	1.14	0.10	8.68
Leu	1.02	0.79	0.91	0.16	18.0
Phe + Tyr	1.17	1.05	1.11	0.08	7.64
Try	-	-	-	-	-
Total	1.25	0.87	1.06	0.27	25.3

Table VI. Summary of the amino acid profiles into factors A and B

	Factor A		
	Muscle	Skin	Factor B means
Amino acid composition (Factor B)			
Total essential amino acid	43.5	30.4	37.0
Total non-essential amino acid	44.1	29.1	36.6
Factor A means	43.8	29.8	36.8

Table VII. Summary of the statistical analysis of the data in Tables I, II, III, IV and V

From Table	r_{xy}	r_{xy}^2	R_{xy}	C_A	IFE	Remark
I	0.9934	0.99	1.36	0.10(10%)	0.90 (90%)	*
II(pI)	0.9382	0.88	1.35	0.35(35%)	0.65(65%)	*
III	0.7658	0.59	0.20	0.89(89%)	0.11(11%)	*
IV	0.5386	0.29	0.24	0.84(84%)	0.16(16%)	NS[a]
V	0.5899	0.35	0.39	0.81(81%)	0.19(19%)	NS

[a]Not significant. *, significant at $r_{=0.05}$ at n -2 degrees of freedom.