

IMPROVEMENT THE PROPERTIES OF YOGHURT USING LEGUMES TO THERAPY PROTEIN ENERGY MALNUTRITION

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ABSTRACT: *The aim of this study was to improve the properties of yoghurt using different kinds of legumes such as chickpea (0.0, 3.0 and 6.0 % w/w), kidney bean (0.0, 3.0 and 6.0 % w/w) and cowpea (0.0, 3.0 and 6.0 % w/w) in order to therapy protein energy malnutrition in children. Chemical composition, pH, texture, viscosity, and sensory evaluation were determined for all treatments. Results showed that total protein increased in yoghurt treatments as a result of adding legumes compared with control. Yoghurt produced using 6% kidney beans (T2b) had highest total protein compared with other treatments. Yoghurt produced using cowpea 6% (T3a) had the higher total solids. Yoghurt produced with 6% kidney bean (T2b) and produced using 6% cowpea had the lowest fat / dry matter as compared with other treatment. There were no significant differences in pH values among all treatments. Also results showed that for all treatments as the rate of legumes increased the viscosity value also increased. The highest viscosity level was recorded with adding cowpea at rate 6% (T3b). Texture parameters hardness, adhesiveness, cohesiveness, springiness, chewiness and gumminess increased with adding all type of legumes. Yoghurt produced with 6% kidney bean and chickpea 3% had improved rheological and organoleptic Properties compared with other treatments.*

Key words: *Malnutrition, Protein-energy malnutrition, legumes and yoghurt.*

INTRODUCTION

Malnutrition is globally the most important risk factor for illnesses and death, affecting especially hundreds of millions of pregnant women and young children. It is currently the leading cause of global burden of disease (Ezzati *et al.*, 2002). The World Health Organization has defined malnutrition as 'the cellular disparity amid the supply of energy, nutrients and the body's demand for them to ascertain maintenance, growth and specific functions (Anstead *et al.*, 2001; Dean *et al.*, 2003 and Rizwana *et al.*, 2015).

Protein-energy malnutrition is defined as a range of pathological conditions arising from a lack of adequate protein and calories (Ernest *et al.*, 2013). It is a problem in many developing countries, of which African countries are mostly affected, in children between the ages of 6 months and 5 years. This type of malnutrition presents itself in the form of

kwashiorkor, marasmus and marasmic-kwashiorkor (Ernest *et al.*, 2013 and Une & Gupta, 2013). The dietary management of moderate acute malnutrition should normally be based on the optimal use of locally available nutrient-dense foods to improve the nutritional status of children and prevent them from becoming severely acutely malnourished (Ashworth and Ferguson, 2009).

Legumes are higher in protein than any other food plant with values ranging from 17% to 31% and the average about 25%. Legumes are close to animal meat in quality and low-cost dietary vegetable proteins and minerals when compared with animal products. (Adeyeba, 2014). Legumes are featured by their high nutritional value and can be recognized and labeled as both a source of vegetables and a source of protein. They are especially characterized as a good source of protein and are compared with

meat, fish and eggs. They are a relatively cheaper than meat products which make them a good protein-rich alternative regardless of socioeconomic status. (Abrahamson *et al.*, 2006).

Legumes can be a valuable source of energy. The high carbohydrate content contributes a great deal to the energy supply of pulses. The energy content of most pulses has been found to be between 300 and 540 Kcal / 100 g. Energy is required for all metabolic processes. The energy of Pulses comes from the nutrient supply of protein, fat and carbohydrate for example Cowpeas 340 kcal/100 g and Chickpeas 347 kcal/100 g (Reddy *et al.*, 1985 and Oke *et al.*, 1995).

Yoghurt is defined by the Codex Alimentations of 2003 as a coagulated milk product that results from the fermentation of milk by *streptococcus thermophilus* and *lactobacillus delbrueckii spp. bulgaricus* (CODEX STAN 243-2003). As starter culture for yoghurt production, lactic acid bacterial species display symbiotic relations during their growth in milk medium (Tamime and Robinson, 1999). Yoghurt is a nutrient-dense food that meets a wide variety of nutritional needs at for everyone. It is a rich source of milk proteins, carbohydrate, minerals such as calcium and phosphorous, and vitamins such as riboflavin (B2), thiamin (B1), coalmine (B12), folate (B9), niacin (B3) and vitamin A (Mckinley, 2005).

The objective of this study was to use formulate energy dense yoghurt based weaning food rich in nutrition by supplementation with cow pea, kidney bean and chickpea powder and evaluate the effect of legumes powder addition on microbiological, physicochemical, and sensory of yoghurt.

MATERIALS AND METHODS

1. Materials

Fresh full-fat cow milk was obtained

from local market. The composition of raw milk was as follows: fat 3%, protein 2.90%, total solids 11.9%, acidity was 0.19% and pH was 6.64.

Starter cultures LAB. (Express 0.2, thermophilic yoghurt culture Yo-Flex Express), consisting of *Lactobacillus delbrueckii ssp. bulgaricus* and *Streptococcus thermophilus* for manufacture of yoghurt, was obtained from Chr. Hansen's Lab., Denmark.

Legumes: Three types of legumes were used:

Chickpea *Cicerarie thinum*, (protein 20.47%, fat 6.04%, fiber 3.5% and carbohydrate 27.42%) , kidney bean *Phaseolus vulgaris L.*, (protein 28.7%, fiber 6.4 % and fat 0.5 % and carbohydrate 22.8 %) and Cowpea *Vignaun guiculata*, (protein 24 %, fat 1.5 %, fiber 6.6 % and carbohydrate 27.3 %) were obtained from local market. The seeds were thoroughly cleaned from dust and other extraneous materials prior to use. (Bravo *et al.*, 1999)

2. Methods

2.1 Preparation of legumes: The whole legumes of Chickpea, Cowpea and kidney bean were soaked in distilled water (1:10 w/v) for 24 h at room temperature (25 °C). Hulls were removed manually after soaking the seeds according to El-Beltagy, (1996). Seeds were placed in a Birex pot with distilled water (1:10 w/v), then cooked in a microwave oven (Sumsung 44L-900W) on high for 15 min (the seeds were soft when felt between the fingers). Beans were oven-dried at 80 °C for 24 h to constant weight. The dried seeds were milled into flour using laboratory grinding machine (poly mix PX-MFC 90D, Switzerland) and stored in airtight plastic container at 4 °C until use.

2.2 Manufacture of yoghurt: Fresh full cow's milk was supplemented with 0,

3 and 6% legumes powder respectively which were added individually. The control was full fat milk (3% fat, 13.2% TS) without any additives. Chickpea, cow pea and kidney bean powder were added at 3 and 6 % individually. Yoghurt was manufactured according to the protocol proposed by Tamime and Robinson, (1999).

Individual milk samples were heat treated at 90°C for 10 min, cooled to 42°C and inoculated with yoghurt culture at the rate recommended by suppliers, Incubation was done at 42°C till the pH reaching 4.9. This was followed by fast cooling to 7°C keeping the product at the same temperature overnight to represent fresh samples yoghurt was stored at 7±1°C for 14 days.

3. Methods of analysis:

Sampling: Yoghurt samples were taken when (fresh and at 7 and 14 days).

Chemical analysis:

All samples were analyzed for fat and total protein and dry matter according to A.O.A.C. (2000). And for pH according to Ling (1963).

Viscosity:

Viscosity was measured using oscillatory viscometer (VR 3000M YR viscometers, Spain), using spindle 4 at speed of 200 r.p.m at 10°C.(Lal *et al.*, 2006)

Texture analysis:

Textural properties of yoghurt were evaluated using a texture analyzer (FTC TMS-Pro), USA). Yoghurt samples were evaluated in their cups. Hardness, cohesiveness, springiness and chewiness were evaluated in triplicate as described by Szczesniak *et al.*, (1963) and Bourne, (1978).

Sensory evaluation:

The samples were assessed for colour, flavour, viscosity, taste and overall

acceptability using a nine-point hedonic scale, where 9 indicated “like extremely” and 1 indicated “dislike extremely”. Each panelist was provided with enough privacy to avoid biased assessment (Makanjuola, 2012).

Statistical analysis:

Results were expressed as mean ± standard deviation (SD) and least significant differences (LSD) for replicates and subjected to Costat, 6.4 (1998/2008) that was done to determine the degree of significant among treatments and within storage period.

RESULTS AND DISCUSSION

1. Chemical analysis:

Data in Table (1) present the chemical composition including total protein content (T.P), fat content / dry matter (F / DM) and total solids (T.S) of yoghurt as affected by adding different legumes at different ratio when fresh . The obtained results showed that T.P% increased in yoghurt treatments as a result of adding legumes compared with control. These results are in agreement with Zare *et al.*, (2011) who mentioned that legumes are high alternative sources of protein compared with other types of plants. Yoghurt produced using 6% kidney beans (T2b) had higher T.P. % compared with other yoghurt treatments. This result is in agreement with Rehman & Shah (2004) and Yin *et al.*, (2008) who showed that kidney beans had highest content in energy, proteins, carbohydrates minerals and vitamins of the pulse fraction.

Total solids (T.S.) % increased as the ratio of legumes increased in all treatments compared to control. Yoghurt produced using cowpea (T3a) had the higher T.S% compared with all other treatments.

Results also showed that yoghurt produced using kidney beans 3% and cow pea 6% had the lowest F / DM % compared with other yoghurt treatments and control.

Addition of legumes caused significant differences in F /DM% in all treatments compared with control. Yoghurt produced with 6% kidney bean (T2b) and produce using 6% cow pea had the lowest F /DM % as compared with other treatment.

2. pH values

Data in Table (2) describe the changes

in pH values of control yoghurt and treatments produced using legumes in fresh and during storage. Legumes were not markedly affected pH values in all treatments compared with control when fresh while at the end of storage period pH values were significantly decreased in all treatments.

Table (1): Effect of type and ratio of legumes on chemical properties of set yoghurt

*Treatments	(T.P.)%	(T.S.)%	F\DM%
C1	3.18±0.04	12.07±0.24	25±0.01
T1a	3.94±0.02	14.66±0.06	26±0.01
T1b	4.38±0.04	16.80±0.08	24±0.00
T2a	3.84±0.08	14.53±0.04	23±0.00
T2b	4.55±0.13	16.89±0.03	21±0.01
T3a	3.74±0.02	14.85±0.10	23±0.00
T3b	3.92±0.66	17.03±0.07	21±0.00
LSD	0.61	0.26	0.01

Data are Mean ± S.D., LSD (0.05)

* C: control yoghurt made from full fat cow's milk. T1a: yoghurt made from full fat cow's milk + 3% chickpea.

T1b: yoghurt made from full fat cow's milk + 6% chickpea. T2a: yoghurt made from full fat cow's milk + 3% kidney bean.

T2b: yoghurt made from full fat cow's milk + 6% kidney bean. T3a: yoghurt made from full fat cow's milk + 3% cowpea.

T3b: yoghurt made from full fat cow's milk + 6% cowpea.

Table (2): Effect of type and ratio of legumes on pH values of set yoghurt during storage period

*Treatments	Storage period			Mean	LSD
	Fresh	7 days	14 days		
C1	4.71± 0.01	4.41±0.01	4.32± 0.01	4.48	—
T1a	4.52± 0.01	4.29±0.05	4.20± 0.08	4.33	
T1b	4.60± 0.10	4.41± 0.08	4.30± 0.07	4.46	
T2a	4.55± 0.03	4.33± 0.02	4.18±0.04	4.35	
T2b	4.60± 0.04	4.32± 0.07	4.21±0.05	4.37	
T3a	4.61±0.04	4.32± 0.13	2.65±2.16	3.86	
T3b	4.58±0.01	4.35±0.04	4.17±0.06	4.36	
LSD	—				
Mean	4.61	4.34	4.00		
LSD	0.27				

See legend to table (1) for details.

3. Viscosity

Significant variation was noted in viscosities of yoghurt from different treatments produced by adding legumes. Results in Table (3) showed that the highest viscosity level was recorded with adding cowpea at rate 6% (T3b). For all treatments as the rate of legumes increased the viscosity value also increased. The increase of viscosity may be due to the interaction between the legumes and protein particles thus contributing a strong gel when the concentration was doubled.

4. Texture properties

Rheological properties for foods, such as fermented dairy products, are important in the design of flow processes, quality control, storage and processing and in predicting the texture of food (shaker *et al.*, 2000). The hardness of

yoghurt is directly dependent on its total solids and specifically protein content and the type of proteins. Higher protein content would cause a higher degree of cross-linkage of the gel network, resulting in a much denser and more rigid gel structure (Tamime, 2006). Table (4) showed texture parameters (Hardness, Adhesiveness, Cohesiveness, Springiness, Chewiness and Gumminess) of the different treatment of yoghurt over storage. Texture parameters were increased with adding all type of legumes. Higher concentration of all legumes increased fracturability and firmness in samples when compared with low level and control. These results are agreement with (Sandoval–castilla *et al.*, 2004) who illustrated that legumes flour could be potentially consider as texture improvement ingredient for yoghurt supplementation.

Table (3): Effect of type and ratio of legumes on viscosity value of set yoghurt during storage period

*Treatments	Storage period			Mean	LSD
	Fresh	7 days	14 days		
C1	6789.0±83.44	7022.0±186.68	7359.5±183.14	7056.83	202.75
T1a	8675.0±35.36	9096.0±166.88	9289.5±34.65	9020.17	
T1b	11066.5±265.17	11791.0±9.90	12144.5±0.71	11667.33	
T2a	9284.0±98.99	9962.0±117.38	10287.0±233.35	9844.33	
T2b	12029.5±7.78	12401.0±79.20	12676.0±280.01	12368.83	
T3a	9261.0±84.85	10061.5±259.51	11116.0±182.43	10146.17	
T3b	11666.0±690.14	12319.0±596.80	12814.5±180.31	12266.50	
LSD	—				
Mean	9824.43	10378.93	10812.43		
LSD	143.37				

See legend to table (1) for details.

Table (4): Effect of type and ratio of legumes on Texture properties of set yoghurt during storage period

Texture properties	Storage period	*Treatments							Mean	LSD
		C	T1a	T1b	T2a	T2b	T3a	T3b		
Hardness (g)	Fresh	98.50± 4.95	135.00± 14.14	215.00± 7.07	159.50± 7.78	230.00± 21.21	145.00± 0.00	247.50± 9.19	175.79	6.88
	7 days	111.00± 4.24	155.00± 14.14	265.50± 13.44	182.50± 21.92	254.50± 13.44	170.50± 4.95	276.00± 15.56	202.14	
	14 days	122.50± 2.12	177.00± 14.14	309.00± 15.56	201.00± 12.73	288.00± 14.14	189.50± 3.54	305.00± 0.00	227.43	
	Mean	110.67	155.67	263.17	181.00	257.50	168.33	276.17		
LSD		9.72								
Adhesiveness (g.mm)	Fresh	65.50± 3.54	113.00± 1.41	141.50± 4.95	119.50± 3.54	129.50± 6.36	94.50± 4.95	144.50± 13.44	115.43	4.86
	7 days	73.00± 2.83	121.50± 3.54	160.50± 6.36	125.50± 9.19	141.50± 10.61	103.00± 1.41	161.50± 23.33	126.64	
	14 days	75.00± 4.24	127.50± 6.36	169.50± 6.36	138.50± 9.19	159.00± 4.24	105.00± 1.41	175.00± 14.14	135.64	
	Mean	71.17	120.67	1157.17	127.83	143.33	100.83	160.33		
LSD		6.87								
Cohesiveness	Fresh	0.31± 0.02	0.39± 0.05	0.43± 0.02	0.44± 0.01	0.45± 0.04	0.42± 0.00	0.45± 0.01	0.41	—
	7 days	0.33± 0.02	0.42± 0.01	0.44± 0.02	0.45± 0.00	0.46± 0.04	0.44± 0.03	0.46± 0.02	0.43	
	14 days	0.34± 0.01	0.43± 0.01	0.45± 0.02	0.45± 0.01	0.45± 0.06	0.44± 0.01	0.46± 0.01	0.43	
	Mean	0.32	0.41	0.44	0.45	0.45	0.43	0.45		
LSD		—								
Springiness (m.m)	Fresh	0.53± 0.01	0.70± 0.06	0.80± 0.01	0.91± 0.02	0.95± 0.02	0.75± 0.01	0.88± 0.04	0.79	—
	7 days	0.60± 0.02	0.73± 0.07	0.82± 0.01	0.88± 0.06	0.97± 0.02	0.75± 0.01	0.89± 0.04	0.80	
	14 days	0.63± 0.02	0.76± 0.05	0.84± 0.02	0.88± 0.08	0.98± 0.02	0.77± 0.04	0.95±0.01	0.83	
	Mean	0.58	0.73	0.82	0.89	0.96	0.75	0.91		
LSD		—								
Chewiness (mJ)	Fresh	17.31± 0.45	35.92± 4.15	73.03± 0.05	62.79± 3.51	96.35± 0.93	45.37± 0.43	96.94± 6.73	61.10	2.63
	7 days	22.65± 1.41	47.32± 1.86	94.59± 1.81	71.99± 4.03	111.55± 5.24	56.37± 6.31	111.65± 6.41	73.73	
	14 days	26.67± 1.56	56.65± 1.76	114.68± 3.22	78.95± 4.50	126.08± 12.42	63.88± 6.19	133.25± 2.11	85.74	
	Mean	22.21	46.63	94.10	71.24	111.32	55.21	113.95		
LSD		3.72								
Gumminess (N)	Fresh	31.55± 1.55	51.63± 1.24	91.30± 1.56	69.36± 2.26	101.98± 1.31	60.90± 0.00	110.11± 2.34	73.83	2.48
	7 days	37.82± 1.34	65.00± 3.75	115.35± 0.21	82.13± 9.86	115.56± 2.88	75.09± 7.00	125.42± 1.22	88.05	
	14 days	41.60± 2.55	75.28± 7.26	137.34± 0.37	90.36± 2.88	129.20± 9.93	83.41± 4.24	140.30± 4.31	99.64	
	Mean	36.99	63.97	114.66	80.61	115.58	73.13	125.27		
LSD		3.51								

See legend to table (1) for details.

Improvement the properties of yoghurt using legumes to therapy

5. Organoleptic Properties

The results of the evaluation of different yoghurt treatments through 14 days of the storage periods are shown in Table (5). Results indicated that addition of legumes improved organoleptic properties as compared with control yoghurt. Yoghurt produced using legumes increase score of colour in all treatments compared with control. Yoghurt fortified with kidney bean 6% had

the higher colour compared with other treatments. There was no significant difference in colour score in all treatments during storage period. Addition of legumes at different ratio improved the color, flavor, taste, viscosity and overall acceptation of yoghurt compared with control. For all level, as the storage period advanced the scoring point of organoleptic properties gradually decreased.

Table (5): Effect of type and ratio of legumes on sensory evaluation of set yoghurt during storage period

Organoleptic Properties	Storage Period	C1	T1a	T1b	T2a	T2b	T3a	T3b	Mean	LSD
Colour	Fresh	6.10±0.74	8.40±0.84	8.20±0.92	8.00±0.67	8.30±0.95	8.00±0.94	8.10±0.74	7.87±1.09	----
	7 days	5.90±0.88	8.10±0.88	7.90±0.88	7.90±0.99	8.20±0.92	8.40±0.84	8.00±1.05	7.77±1.18	
	14 days	5.91±0.83	7.80±1.14	8.20±0.92	8.20±0.79	8.10±0.57	7.90±0.99	8.10±0.74	7.72±1.15	
Mean		5.97±1.19	8.10±0.96	8.10±0.93	8.03±0.83	8.20±0.86	8.10±0.79	8.07±0.93		
LSD		0.68								
Taste	Fresh	5.10±0.88	8.80±0.42	8.50±0.85	8.80±0.42	8.80±0.42	8.10±0.74	8.30±0.95	8.06±1.41	----
	7 days	5.00±0.94	8.50±0.71	8.50±0.71	8.20±0.79	8.60±0.52	8.60±0.70	8.70±0.48	8.01±1.42	
	14 days	5.00±0.89	8.50±0.71	8.50±0.85	8.50±0.71	8.90±0.32	8.50±0.71	8.30±0.95	7.99±1.49	
Mean		5.03±1.75	8.60±0.62	8.50±0.73	8.50±0.77	8.77±0.45	8.40±0.68	8.43±0.77		
LSD		0.56								
Flavor	Fresh	5.60±1.07	8.40±0.52	8.40±0.70	8.10±0.88	8.60±0.52	8.30±0.82	7.90±0.74	7.90±1.22	----
	7 days	5.70±1.16	8.00±0.67	7.90±0.74	8.30±0.82	8.40±0.52	8.30±0.67	8.20±0.63	7.83±1.15	
	14 days	5.18±1.17	8.00±0.67	8.40±0.70	8.00±0.67	8.30±0.48	8.00±0.67	7.90±0.74	7.65±1.30	
Mean		5.48±1.46	8.13±0.63	8.23±0.71	8.13±0.77	8.43±0.61	8.20±0.65	8.00±0.64		
LSD		0.59								
Viscosity	Fresh	4.20±0.79	8.80±0.42	8.10±0.74	8.10±0.74	8.50±0.53	8.40±0.70	8.00±0.67	7.73±1.61	----
	7 days	4.30±0.82	8.70±0.48	8.20±0.79	8.40±0.70	8.60±0.52	8.60±0.70	8.50±0.71	7.90±1.63	
	14 days	4.09±0.83	8.20±0.79	8.10±0.74	8.40±0.70	8.20±0.42	8.50±0.71	8.00±0.67	7.59±1.66	
Mean		4.19±1.88	8.57±0.63	8.13±0.73	8.30±0.71	8.43±0.51	8.50±0.63	8.17±0.69		
LSD		0.53								
overall acceptability	Fresh	5.50±0.85	8.60±0.52	8.60±0.52	8.60±0.70	8.50±0.53	8.30±0.82	8.30±0.67	8.06±1.24	----
	7 days	5.60±0.97	8.20±0.63	8.10±0.74	8.20±0.79	8.40±0.52	8.60±0.52	8.30±0.67	7.91±1.18	
	14 days	5.55±1.04	7.90±0.74	8.60±0.52	8.40±0.70	8.50±0.71	8.10±0.74	8.30±0.67	7.87±1.25	
Mean		5.55±1.51	8.23±0.68	8.43±0.68	8.40±0.68	8.47±0.57	8.33±0.68	8.30±0.65		
LSD		0.54								

See legend to table (1) for details.

Conclusion

The obtained results suggest possibility of making a good quality high protein and calorie yoghurt with the use of legumes. Results also indicated that 6% kidney bean and 3% chickpea flour may be useful ingredient for production of yoghurt without adversely effect on the properties of the product.

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تحسين خصائص اليوجورت باستخدام البقوليات لعلاج سوء التغذية الناتج عن نقص البروتين في الاطفال

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الملخص العربي

تهدف هذه الدراسة الى تحسين خصائص اليوجورت باستخدام أنواع مختلفة من البقوليات بنسب مئوية مختلفة مثل إضافة الحمص (٦، ٣، ٠)، الفاصوليا (٦، ٣، ٠)، اللوبيا (٦، ٣، ٠) وزن/وزن وذلك لعلاج سوء التغذية الناتج عن نقص البروتين في الأطفال. تم تقييم التركيب الكيميائي، درجة الحموضة، اللزوجة، الخواص الريولوجية و التقييم الحسي لجميع المعاملات. اوضحت النتائج زيادة نسبة البروتين في المعاملات باضافة البقوليات إلي اليوجورت مقارنة بالكنترول. اليوجورت المنتج باضافه ٦% من الفاصوليا يحتوي علي اعلي نسبة بروتين مقارنة بباقي المعاملات. اليوجورت المنتج باضافه ٦% من اللوبيا يحتوي علي اعلي نسبة مواد جافة مقارنة بباقي المعاملات، اليوجورت المنتج باضافه ٦% من الفاصوليا و٦% من اللوبيا يحتويان علي اقل نسبة دهن / المادة جافة مقارنة بباقي المعاملات. أيضا اوضحت النتائج ارتفاع اللزوجة باضافة اللوبيا بنسبة ٦%. وزيادة نسبة اللزوجة بزيادة النسبة المضافة من البقوليات. لا يوجد فروق معنوية في قيمة الـ pH بين كل المعاملات. الخواص الريولوجية مثل الصلابة، اللزوجة والمرونة، المضغية والتماسك زادت باضافة كل انواع البقوليات. اليوجورت المنتج باضافه ٦% من الفاصوليا و٣% من الحمص كان له أفضل الخواص الريولوجية و الحسية مقارنة بباقي المعاملات.

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