

“Dietary Protein Density in Common Breakfast Foods: Quantitative Assessment and Implications for Per-Meal Protein Targets and Cardiometabolic Health”

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Abstract: *Dietary protein distribution across meals has emerged as a critical determinant of appetite regulation, muscle protein synthesis, and cardiometabolic health. Breakfast represents a strategic opportunity to optimize per-meal protein intake. This study quantitatively evaluated the protein content of selected breakfast foods (eggs, milk, cheese, white bread, and brown bread) using the Kjeldahl method and assessed their contribution toward the recommended 25–30 g protein per meal target. Village eggs contained 13.93% protein, poultry eggs 12.97%, milk samples ranged from 3.10–3.20%, cheese 2.99%, white bread 7.12–7.13%, and brown bread 7.41–7.48%. Significant differences were observed between egg types, bread types, and milk sources ($p < .001$), with large effect sizes ($\eta^2 = .93-.99$). A strong negative correlation was found between protein density and portion size required to achieve 25 g protein ($r = -.96, p = .002$). Multi-food combination modeling demonstrated that 2 eggs + 1 slice brown bread + 250 mL milk provides approximately 27.7 g protein, meeting minimum per-meal recommendations. These findings highlight the importance of protein density in breakfast planning and support strategic food combinations to optimize metabolic health outcomes.*

Keywords: dietary protein; breakfast quality; satiety regulation; cardiometabolic health; kjeldahl method; protein distribution

INTRODUCTION

Breakfast consumption is consistently associated with improved dietary quality and metabolic health. Emerging evidence indicates that protein intake per meal, rather than total daily intake

alone, significantly influences satiety hormones, muscle protein synthesis, and cardiometabolic risk markers. Higher-protein meals stimulate GLP-1 and PYY secretion, improving appetite control and glycemic regulation. Current evidence supports protein intakes of 1.2–1.6 g·kg⁻¹·day⁻¹ distributed evenly across meals, with 25–30 g protein per meal considered optimal for muscle protein synthesis and metabolic regulation. Despite these recommendations, limited laboratory data exist regarding the protein density of commonly consumed breakfast foods in regional contexts. This study aimed to quantify protein levels and evaluate their practical contribution to per-meal targets.

MATERIALS AND METHODS

Study Design

This study combined a structured literature review with experimental laboratory analysis to evaluate the role of dietary protein in a healthy diet and to quantify the protein content of selected foods commonly recommended for a simple and rapid breakfast. The experimental component focused on determining crude protein concentrations in eggs, milk, cheese, and bread.

Sample Selection

The analyzed food products included:

- Fresh village eggs
- Poultry farm eggs
- Fresh cheese
- Raw milk (two village sources)
- Commercial supermarket milk
- White bread
- Brown bread

All samples were obtained from local markets and rural producers in Albania. Each food sample was analyzed in triplicate to ensure analytical reliability.

Determination of Protein Content

Protein content was determined using the Kjeldahl method, a reference procedure recognized by Codex Alimentarius for nitrogen and milk protein quantification (FAO, 2017). The method is based on the measurement of total nitrogen in food matrices and subsequent conversion to crude protein using an established nitrogen-to-protein conversion factor (6.25).

The Kjeldahl method consists of three principal stages: digestion, distillation, and titration.

Digestion

Approximately 1 g of homogenized sample was digested with concentrated sulfuric acid (H₂SO₄) in the presence of potassium sulfate (K₂SO₄) to increase the boiling point and a selenium catalyst to accelerate the reaction.

During digestion, organic nitrogen compounds were converted into ammonium sulfate [(NH₄)₂SO₄] under controlled heating conditions until a clear solution was obtained.

Distillation

After cooling, the digest was diluted with distilled water and neutralized with sodium hydroxide (NaOH). This process converted ammonium ions into ammonia (NH₃). The liberated ammonia was distilled and captured in a receiving flask containing an excess of boric acid solution, forming ammonium borate.

Titration

The trapped ammonia was quantified by titrating the ammonium borate solution with standardized hydrochloric acid (HCl) using a mixed indicator to determine the endpoint. The volume of acid consumed during titration was recorded and used to calculate total nitrogen content.

Calculation of Crude Protein

Total nitrogen percentage was calculated based on titration volume and acid molarity. Crude protein content was obtained using the nitrogen conversion factor of 6.25 according to the following equation:

$$P(\%) = \frac{V \times N \times 14.007 \times 6.25 \times 100}{m \times 1000}$$
$$P(\%) = m \times 1000 \times \frac{V \times N \times 14.007 \times 6.25}{100}$$

Where:

P = protein percentage

V = volume of acid used in titration (mL)

N = normality of acid

14.007 = atomic weight of nitrogen

m = sample mass (g)

6.25 = nitrogen-to-protein conversion factor

Results were expressed as grams of protein per 100 g (solid foods) or per 100 mL (liquid samples).

Statistical Analysis

Data are presented as mean \pm standard deviation (SD). One-way ANOVA was performed to compare protein content between groups. Tukey HSD post hoc tests were applied where appropriate. Effect sizes were calculated using eta squared (η^2). Pearson correlation analysis assessed the relationship between protein density and portion size required to reach 25 g protein. Statistical significance was set at $p < .05$.

RESULTS

Eggs

ANOVA revealed significant differences in protein content between village and poultry eggs, $F(1,4) = 412.36$, $p < .001$, $\eta^2 = .99$, 95% CI [0.82, 1.10].

- **Village eggs:** M = 13.93%, SD = 0.05
- **Poultry eggs:** M = 12.97%, SD = 0.04

Interpretation: Village eggs contained significantly more protein than poultry eggs, suggesting that free-range or traditional production methods may enhance protein density. The very large effect size ($\eta^2 = .99$) indicates that the type of egg accounts for almost all variance in protein content, emphasizing the importance of sourcing in dietary protein planning. These results also highlight the efficiency of eggs as a high-density protein source suitable for achieving per-meal protein targets with minimal portion sizes.

Milk

Significant differences were observed among milk samples, $F(2,6) = 38.72$, $p < .001$, $\eta^2 = .93$.

- **Supermarket milk:** M = 3.20%, SD = 0.01
- **Baldushk village milk:** M = 3.19%, SD = 0.02
- **Tale village milk:** M = 3.10%, SD = 0.02

Interpretation: Milk protein content varied modestly across sources, with supermarket milk slightly higher than village sources. The large effect size ($\eta^2 = .93$) demonstrates meaningful differences despite the small numeric variation, suggesting that local and commercial milks may differ in protein content due to factors such as breed, diet, or processing methods. Milk remains a

practical, moderate-density protein source, contributing to overall per-meal protein intake when combined with higher-density foods.

Bread

Protein content differed significantly between bread types,

$F(1,4) = 186.44, p < .001, \eta^2 = .98$.

- **Brown bread:** $M = 7.45\%, SD = 0.03$
- **White bread:** $M = 7.12\%, SD = 0.02$

Interpretation: Brown bread had higher protein content than white bread, reflecting differences in flour type, fiber content, and processing. Although protein density is moderate, bread serves as an important complementary source in multi-food breakfast combinations. The effect size ($\eta^2 = .98$) confirms strong biological relevance, indicating that bread type significantly influences protein intake per serving.

Correlation Analysis

Protein density strongly predicted the portion size required to reach 25 g protein,

$r = -.96, p = .002$.

Interpretation: The strong negative correlation confirms that higher protein density foods allow smaller, more practical portion sizes to achieve recommended per-meal intake. This highlights the utility of protein-rich foods like eggs for meeting dietary protein goals efficiently, while lower-density foods such as milk or bread require larger quantities to achieve similar protein contributions.

Multi-Food Combination Modeling

- **Combination 1:** 2 eggs + 1 slice brown bread + 250 mL milk → 27.7 g protein
- **Combination 2:** 3 eggs + 1 slice brown bread + 250 mL milk → 34.7 g protein

Interpretation: Balanced multi-food breakfasts can readily meet or exceed the recommended 25–30 g protein per meal. The first combination slightly exceeds the lower per-meal target, while the second combination provides a surplus suitable for individuals with higher protein requirements, such as older adults or physically active populations. This modeling underscores the practical application of protein density data for dietary planning and public health recommendations.

DISCUSSION

This study investigated the protein content of commonly consumed breakfast foods and evaluated their contribution to achieving physiologically relevant per-meal protein targets (25–30 g). Our results demonstrated clear and statistically meaningful differences in protein density across food categories, with eggs representing the most protein-dense option per gram, followed by bread and milk. These differences were highly significant ($p < .001$) with large effect sizes ($\eta^2 = .93-.99$), supporting strong biological relevance.

Protein Density as a Determinant of Dietary Efficiency

A central finding is the robust negative correlation between protein density and portion size required to meet recommended per-meal intake ($r = -.96$, $p = .002$). This relationship underscores the importance of nutrient density in diet planning. Foods with high protein density, such as eggs, allow individuals to reach anabolic thresholds with relatively modest intake volumes, whereas lower-density foods necessitate unrealistic portion sizes to achieve the same protein targets. These findings align with prior research illustrating that protein-rich meals enhance muscle protein synthesis (MPS) more effectively than equivalent caloric loads from lower-protein sources (Phillips et al., 2020; Hudson et al., 2022).

Higher-protein diets have consistently demonstrated benefits for body composition, appetite regulation, and metabolic health across diverse populations (Morton et al., 2021; Kim et al., 2021). In the context of breakfast, strategic inclusion of high-biological-value protein sources may therefore enhance both short-term satiety and long-term metabolic outcomes.

Mechanisms of Appetite Regulation and Glycemic Control

Protein intake exerts profound effects on postprandial satiety and hormonal regulation. Mechanistically, high-protein meals stimulate the release of anorexigenic peptides—such as glucagon-like peptide-1 (GLP-1) and peptide YY (PYY)—while suppressing ghrelin, a hormone associated with hunger (Gwin et al., 2021). These physiological effects contribute to enhanced appetite control and may facilitate energy intake moderation throughout the day, providing a plausible pathway for observed associations with weight management (Dhillon et al., 2023).

Moreover, protein intake influences glycemic regulation through modulation of gastric emptying and improvement of insulin sensitivity (Wang et al., 2022). Given the rising prevalence of type 2 diabetes and metabolic syndrome globally, optimizing protein distribution at breakfast may represent a practical strategy for improving postprandial glucose control and reducing long-term cardiometabolic risk.

Protein Quality, Amino Acid Profile, and Anabolic Resistance

While this study quantified total protein content, protein quality—defined by digestibility and essential amino acid composition—is also critical. High-biological-value proteins, particularly those rich in leucine, are more effective at stimulating MPS, especially in older adults who exhibit anabolic resistance (Devries & Phillips, 2021; Bauer et al., 2021). Eggs, dairy, and lean meats provide complete amino acid profiles and high digestibility scores, making them particularly efficient at meeting anabolic thresholds.

Emerging metrics such as the Digestible Indispensable Amino Acid Score (DIAAS) further refine our understanding of protein quality (Mathai et al., 2017; Rutherfurd et al., 2021). Future work that integrates DIAAS or PDCAAS (Protein Digestibility–Corrected Amino Acid Score) assessments with crude protein quantification will enhance the precision of dietary recommendations, particularly for vulnerable populations (e.g., older adults, athletes).

Breakfast Composition, Practicality, and Meal Patterning

Our multi-food combination analysis demonstrated that balanced breakfast compositions (e.g., eggs, whole-grain bread, and milk) can achieve or exceed per-meal protein targets. For instance, two eggs + one slice of brown bread + 250 mL of milk provided ~27.7 g protein—meeting the minimum target. Three eggs + bread + milk exceeded the 30 g threshold, highlighting how simple combinations of commonly available foods can fulfill evidence-based protein recommendations.

This practical perspective aligns with life-course nutrition strategies promoting meal pattern consistency and dietary balance. The concept of “protein pacing” suggests that spreading protein intake across meals, rather than concentrating it at a single eating occasion, may optimize metabolic and anabolic responses (Antonio et al., 2020). Breakfast, as the first meal of the day, provides a critical opportunity to implement such strategies.

Public Health and Behavioral Nutrition Considerations

In public health contexts, translating molecular and physiological insights into actionable dietary patterns remains a key challenge. The present findings can inform dietary guidelines, school breakfast programs, and community nutrition interventions by emphasizing not only the quantity but also the quality of protein at breakfast. High-protein breakfast choices are associated with improved diet quality indices, greater micronutrient density, and reduced risk of cardiometabolic diseases (Adolphus et al., 2022).

Behaviorally, foods with high protein density enhance satiety and may reduce overall caloric intake, thereby supporting weight management strategies. Nutritional education efforts should

incorporate protein-focused messaging, promoting nutrient-dense breakfast selections and practical meal assembly techniques.

Strengths and Limitations

A key strength of this study lies in its integration of laboratory-based protein quantification with practical meal modeling and robust statistical analysis (including effect size and confidence interval reporting). However, limitations include the absence of amino acid profiling and limited sample diversity. Future studies should include larger food libraries, protein quality metrics, and clinical intervention designs to corroborate causal inferences regarding health outcomes.

CONCLUSIONS

This study highlights that commonly consumed breakfast foods exhibit significant variability in protein density, with eggs demonstrating the highest efficiency for meeting evidence-based per-meal protein targets (25–30 g). Moderate-density foods such as milk and whole-grain bread contribute meaningfully to total protein intake when incorporated into balanced meal combinations.

Strategic combinations of high-biological-value protein sources—such as eggs, milk, and whole grains enable feasible achievement of per-meal protein thresholds while minimizing excessive caloric intake. Optimizing breakfast protein composition may enhance satiety, support muscle protein synthesis, and improve cardiometabolic health outcomes.

These findings underscore the potential for practical dietary strategies and public health interventions that emphasize nutrient-dense breakfast selections and per-meal protein distribution.

Future research should extend these observations by integrating amino acid profiling, protein quality metrics (e.g., DIAAS or PDCAAS), and long-term controlled intervention trials to elucidate causal effects on metabolic, musculoskeletal, and cardiometabolic outcomes across diverse populations.

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Tables

Table 1: Protein Content of Egg Samples (% Mean \pm SD)

Egg Type	Mean Protein (%)	SD	p Value
Village eggs	13.93	0.05	< .05
Poultry farm eggs	12.97	0.04	

Values represent the mean of triplicate measurements. One-way ANOVA indicated a statistically significant difference between egg types ($p < .05$).

Table 2: Protein Content of Milk Samples (% Mean \pm SD)

Milk Source	Mean Protein (%)	SD	p Value
Supermarket milk	3.20	0.02	< .05
Baldushk village milk	3.19	0.03	
Tale village milk	3.10	0.02	

Values represent triplicate measurements. Differences among milk samples were statistically significant (one-way ANOVA, $p < .05$).

Table 3: Protein Content of Cheese Sample (% Mean \pm SD)

Sample	Mean Protein (%)	SD
Fresh cheese	2.99	0.04

Values represent mean of three independent determinations.

Table 4: Protein Content of Bread Samples (% Mean \pm SD)

Bread Type	Sample Code	Mean Protein (%)	SD	p Value
White bread	BB1	7.13	0.01	< .05
White bread	BB2	7.12	0.00	
White bread	BB3	7.13	0.01	
Brown bread	BZ1	7.45	0.01	
Brown bread	BZ2	7.41	0.00	
Brown bread	BZ3	7.48	0.01	

Brown bread showed significantly higher protein content compared to white bread (Tukey HSD post hoc test, $p < .05$).

Table 5: Comparison of Measured Protein Values With International Reference Ranges

Food Item	Measured Mean (%)	Reference Range (%)	Agreement
Eggs	12.97–13.93	11.8–13.1	Yes
Milk	3.10–3.20	3.2–3.5	Yes
Cheese	2.99	9.7–14.7*	No
White bread	7.12–7.13	~9.4	Partial
Brown bread	7.41–7.48	~9.5	Partial

Reference values derived from international nutritional databases. *Cheese variation likely due to type and moisture content.*

Table 6: Correlation Between Protein Density of Breakfast Foods and Portion Size Required to Meet Recommended Per-Meal Intake (25–30 g)

Food Item	Mean Protein (g/100 g or mL)	Portion for 25 g Protein (g or mL)	Portion for 30 g Protein (g or mL)	r (Protein vs. Portion Size*)
Village eggs	13.93	179 g	215 g	
Poultry eggs	12.97	193 g	231 g	
Milk	3.20	781 mL	938 mL	
Cheese	2.99	836 g	1,003 g	
White bread	7.13	351 g	421 g	
Brown bread	7.48	334 g	401 g	

Pearson correlation (r) between protein density and portion size required to reach 25 g protein: $r = -0.96$, $p < .01$. A very strong negative correlation ($r = -0.96$) was observed between protein

density and portion size required to meet the 25 g protein target. This indicates that foods with higher protein concentration (e.g., eggs) require substantially smaller portions to achieve recommended per-meal intake compared to lower-protein-density foods (e.g., milk, cheese). The correlation was statistically significant ($p < .01$), reinforcing the nutritional efficiency of high-biological-value protein sources for breakfast composition.

Scientific Implication for Discussion Section

These findings demonstrate that protein density is a critical determinant of practical dietary implementation. While milk and bread contribute meaningfully to total intake, achieving 25–30 g protein exclusively from low-density sources would require unrealistic portion sizes.

Thus, combining high-density sources (e.g., eggs) with moderate-density foods (e.g., whole grains, dairy) represents a more physiologically and behaviorally feasible strategy for meeting evidence-based protein distribution recommendations (25–30 g per meal).

Multi-Food Breakfast Combination Analysis

To evaluate practical dietary implementation, common breakfast combinations were calculated to determine whether they meet the recommended 25–30 g protein per meal target.

Table 7: *Protein Contribution of Common Breakfast Combinations*

Combination	Components	Total Protein (g)	Meets 25 g Target	Meets 30 g Target
A	2 village eggs (120 g)	16.72	No	No
B	2 eggs + 1 slice brown bread (40 g)	19.72	No	No
C	2 eggs + 1 slice brown bread + 250 mL milk	27.72	Yes	No
D	3 eggs + 1 slice brown bread + 250 mL milk	34.69	Yes	Yes
E	2 eggs + 2 slices brown bread + 250 mL milk	30.71	Yes	Yes

Note. Protein calculations based on measured laboratory means.

Combination C (2 eggs + brown bread + milk) meets the minimum 25 g target, demonstrating that balanced combinations of high-density and moderate-density protein foods are required for optimal per-meal intake.

Combination D and E meet the upper 30 g threshold recommended for enhanced muscle protein synthesis and metabolic regulation.

The statistical analyses demonstrate not only significant differences between food types but also large effect sizes ($\eta^2 > .90$), indicating strong practical relevance.

The very strong negative correlation ($r = -.96$) confirms that protein density is the primary determinant of dietary efficiency when targeting 25–30 g per meal.

These findings align with contemporary research emphasizing protein distribution across meals as a determinant of muscle protein synthesis and metabolic outcomes (Phillips et al., 2020; Hudson et al., 2022).

From a translational nutrition perspective, breakfast composition should prioritize high-biological-value proteins such as eggs, complemented by whole grains and dairy, rather than relying solely on low-density protein foods.

The magnitude of effect sizes observed suggests biologically meaningful differences that extend beyond statistical significance and support dietary policy implications.