

RIBOFLAVIN CONTENT IN SELECTED TRADITIONAL FOODS FROM BLACK SEA AREA COUNTRIES

H. S. Costa¹, C. Flores¹, A. Sanches-Silva¹, T. G. Albuquerque¹, M. Santos¹, E. Vasilopoulou², A. Trichopoulou^{2,3}, F. D'Antuono⁴, I. Alexieva⁵, O. Hayran⁶, L. Kaprelyants⁷, D. Karpenko⁸, Z. Kilasonia⁹, N. Koval¹⁰, A. L. Stroia¹¹, P. Finglas¹²

¹ Food and Nutrition Department, National Institute of Health Dr. Ricardo Jorge, I.P., Av. Padre Cruz, 1649-016 Lisbon, Portugal; ² Department of Hygiene, Epidemiology & Medical Statistics, Medical School, University of Athens, Greece; ³ Hellenic Health Foundation, Athens, Greece; ⁴ Campus of Food Science, Cesena, University of Bologna, Italy; ⁵ University of Food Technologies, Plovdiv, Bulgaria; ⁶ T C Yeditepe University, Istanbul, Turkey; ⁷ Department of Biochemistry and Microbiology, Odessa National Academy of Food Technologies, Odessa, Ukraine; ⁸ State Educational Institution of the High Professional Education "Moscow State University of Food Productions", Russian Federation; ⁹ Elkana, Biological Farming Association, Tbilisi, Georgia; ¹⁰ UzhNU (Uzhhorod National University), Uzhhorod, Ukraine; ¹¹ The Bucharest Academy of Economic Studies, Romania; ¹² Institute of Food Research, Norwich Research Park, Colney, Norwich, NR47UA, United Kingdom

E-mail: helena.costa@insa.min-saude.pt

BACKGROUND

Riboflavin (vitamin B₂) is an essential water-soluble vitamin present in a wide variety of foods, namely in milk, dairy products, cereal products, meat products and green leafy vegetables. The primary form of the vitamin is an integral component of the coenzymes flavin mononucleotide and flavin-adenine dinucleotide. It is in these bound coenzyme forms that riboflavin functions as a catalyst for redox reactions in numerous metabolic pathways and in energy production. The daily recommended allowance for riboflavin is 1.3 mg/day and 1.1 mg/day, for males and females, respectively [1]. Due to its unquestionable importance in human nutrition, riboflavin was determined in the selected traditional foods analysed in the frame of the European Project BaSeFood (Sustainable exploitation of bioactive components from the Black Sea Area traditional foods) [2].

MATERIALS AND METHODS

Riboflavin determination

The quantitative determination of vitamin B₂ was based on EN 14152:2003 method, an accredited method, by ISO/IEC/17025 [3]. The laboratory also participates successfully in proficiency testing schemes. The samples were extracted after acid hydrolysis followed by dephosphorylation (with enzymatic treatment) and quantified by High Performance Liquid Chromatography with fluorescence detection.

Sample extraction

- Triplicate samples (2 to 10 g) were submitted to acid hydrolysis with HCl 0.1 M for 30 min in an autoclave at 121 °C
- After cooling at room temperature, pH value was adjusted to 4.5 with sodium acetate 2.5 M and samples were submitted to an overnight enzymatic treatment at 37 °C: takadiastase (0.25 g/sample) and β-amylase (0.05 g/sample) (Figure 1)
- After cooling at room temperature, the solutions were diluted to 100 mL with ultra pure water and filtered with 150 mm Ø filter paper (Figure 1)
- Extracts were filtered again through a 0.45 µm PET syringe filter

Chromatographic conditions

- ✓ Equipment: HPLC Waters 2695 Separations Module
- ✓ Detector: 2475 multi-wavelength fluorescence
- ✓ Column: Phenomenex Luna C18 (250 x 4.6 mm I.D., 5 µm particle size)
- ✓ Column temperature: 37 °C
- ✓ Injection volume: 50 µL
- ✓ Flow rate: 1 mL/min
- ✓ Retention time: 10.3 min
- ✓ Detection: λ_{ex} = 422 nm and λ_{em} = 522 nm
- ✓ Mobile phase: 0.05 M acetate buffer + methanol (70:30, v/v)

RESULTS

Figure 2 shows the riboflavin content found in Traditional Foods from Black Sea Area countries. Roasted sunflower seeds presented the highest concentration of riboflavin (0.19 mg/100 g of edible portion) and approximately 42% of the analysed Traditional Foods had contents lower than the limit of quantification (<0.02 mg/100 g of edible portion). Chromatograms of (A) riboflavin standard solution and (B) Traditional food sample are illustrated in Figure 3.

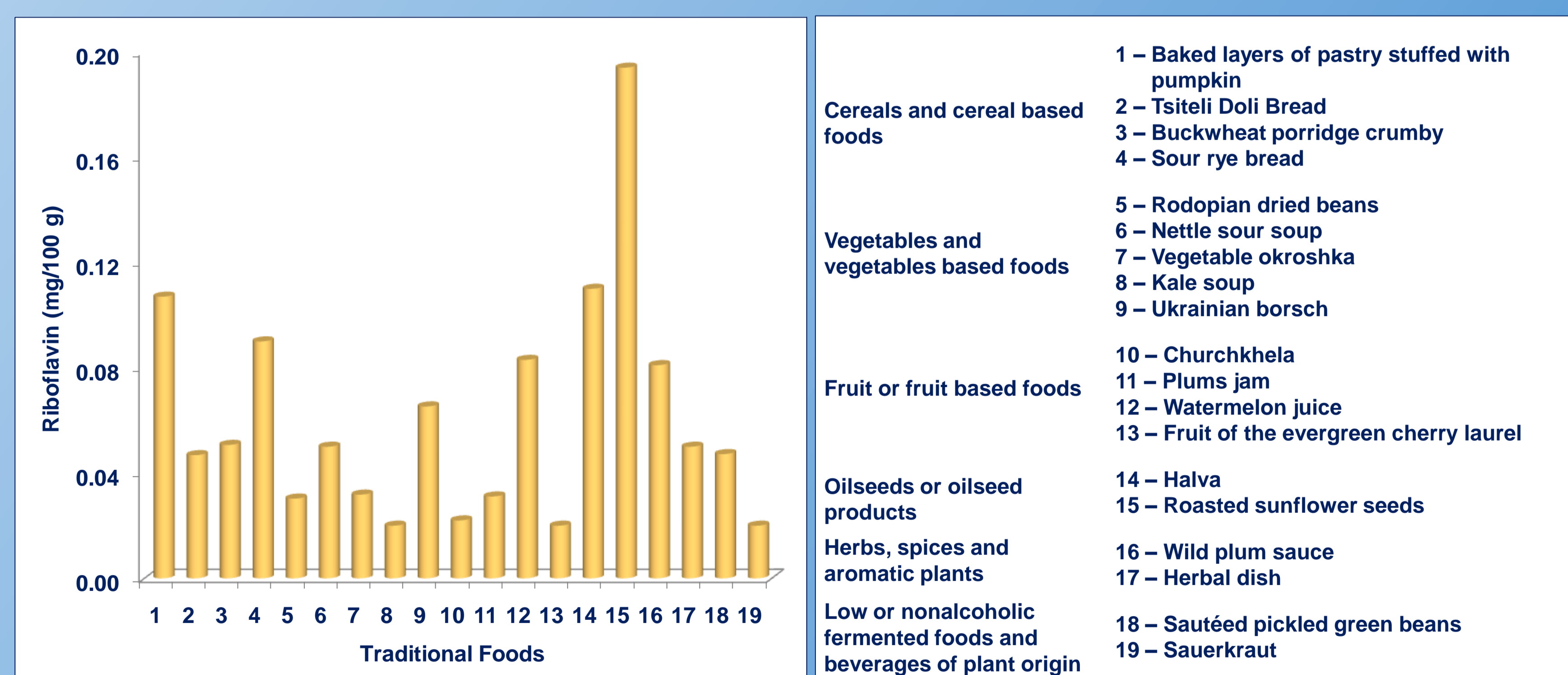


Figure 2. Riboflavin content (mg/100 g of edible portion) in Traditional Foods from Black Sea Area countries.

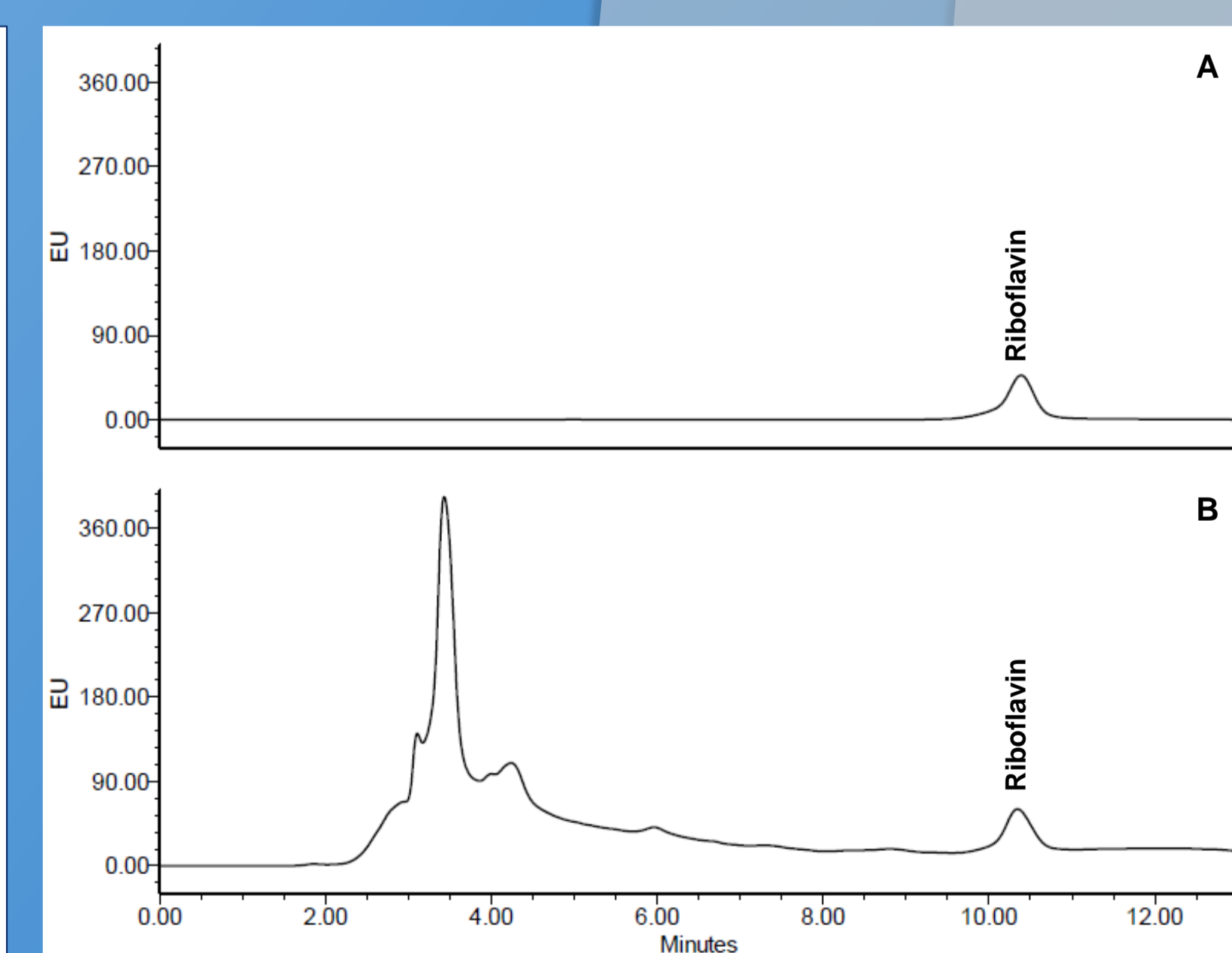


Figure 3. A - Chromatogram of a standard solution of riboflavin (0.080 µg/mL). B - Chromatogram of Churchkhela (0.064 µg/mL).

CONCLUSION

Our results show that 58% of the analysed Traditional Foods from Black Sea Area countries give a contribution to riboflavin dietary intake, especially the groups of oilseeds or oilseed products (roasted sunflower seeds and halva) and cereals and cereal based foods (baked layers of pastry stuffed with pumpkin and sour rye bread).

REFERENCES

- [1] Food and Nutrition Board, Institute of Medicine, National Academies. (2002). Dietary Reference Intakes (DRI's): estimated average intake for groups.
- [2] D'Antuono L.F., Soares Costa H., Sanches-Silva A. (2010). BaSeFood: Sustainable exploitation of bioactive components from the Black Sea Area traditional foods. Nutrition Bulletin, 35, 272-278.
- [3] EN 14152:2003 – Foodstuffs. Determination of vitamin B2 by High Performance Liquid Chromatography.

ACKNOWLEDGEMENTS

The research leading to these results has received funding from the European Community's Seventh Framework Programme (FP7/2007-2013) under grant agreement n.º 227118.



Figure 1. Riboflavin extraction and quantification. (A) and (B) Overnight incubation (C), (D) and (E) Sample filtration (F) Standard solutions of riboflavin (G) HPLC equipment