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INVESTIGACIÓN

Development and consumer acceptance of gluten-free pasta enriched with *Pyropia columbina* seaweed. Physical, textural and nutritional properties

Desarrollo y aceptación por parte del consumidor de pasta sin gluten enriquecida con algas *Pyropia columbina*. Propiedades físicas, texturales y nutricionales

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ABSTRACT

Introduction: Gluten-free pasta is low in protein, and involve a loss of quality in cooking and texture. The objective of this work was to increase the nutritional value of gluten-free pasta through the incorporation of different amounts of *Pyropia columbina* seaweed powder and to analyze their effects on physicochemical and sensory quality.

Methodology: Pasta was made from cassava starch and corn flour (80:20), milk, egg, salt and xanthan gum, with the addition of *Pyropia columbina* seaweed powder (6.0 and 9.0%). Cooking quality (optimal cooking time, water absorption and cooking loss), textural quality (firmness, springiness, chewiness and shear force) and analytical sensory test were analyzed; also, chemical composition, *in vitro* protein digestibility and general acceptance of the chosen formulation in comparison to commercial pasta with similar characteristics, were analyzed. Data obtained of the parameters were analyzed through ANOVA and Fisher's test.

Results: Results showed that pasta enriched with 6.0% seaweed developed lower cooking loss regarding to control, better textural quality and sensory characteristics. This formulation showed a significant increase in protein, ash, total dietary fiber and a decrease in fat content, which caused a decrease in available carbohydrates and caloric value. In addition, an increase in the mean protein digestibility value was obtained, as well as better consumer acceptance.

Conclusions: Addition of *Pyropia columbina* seaweed powder at 6.0% in this gluten-free pasta formulation increased the protein content by 23%, total dietary fiber by 50%, and ash by 150% and developed the characteristics of quality required for pasta. The obtained pasta shows great potential as a functional food and was mostly accepted by consumers.

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Keywords: Seaweed; Food, Fortified; Proteins; Gluten-Free Diet; Consumer Satisfaction.

Entry terms: Fortified pasta.

RESUMEN

Introducción: La pasta sin gluten es baja en proteínas, lo que implica una pérdida de calidad en la cocción y textura. El objetivo de este trabajo fue aumentar el valor nutricional de una pasta sin gluten mediante la incorporación de diferentes cantidades de polvo de alga *Pyropia columbina* y analizar sus efectos sobre la calidad fisicoquímica y sensorial.

Metodología: Las pastas fueron elaboradas a partir de almidón de mandioca y harina de maíz (80:20), leche, huevo, sal y goma xántica, con adición de polvo de alga *Pyropia columbina* (6,0 y 9,0%). Se analizaron calidad de cocción (tiempo óptimo de cocción, absorción de agua y pérdida por cocción), calidad de textura (firmeza, elasticidad, masticabilidad y fuerza de corte) y pruebas sensoriales analíticas; además, se analizó la composición química, la digestibilidad de la proteína *in vitro* y la aceptación general de la formulación elegida comparada con una pasta comercial con similares características Los datos obtenidos de los parámetros se analizaron mediante ANOVA y la prueba de Fisher.

Resultados: Los resultados mostraron que la pasta enriquecida con 6,0% de algas presentó menos pérdida por cocción en comparación con el control, mejor calidad de textura y características sensoriales. Esta formulación mostró un aumento significativo en proteínas, cenizas, fibra dietética total y una disminución en el contenido de grasas, lo que provocó una disminución de los carbohidratos disponibles y del valor calórico. Además, se obtuvo un incremento en el valor medio de la digestibilidad de las proteínas; así como mejor aceptación del consumidor.

Conclusiones: El agregado de 6,0% de polvo de algas *Pyropia columbina* en esta formulación de pasta sin gluten aumentó el contenido de proteína en 23%, fibra dietética total en 50% y cenizas en 150%. La pasta obtenida muestra gran potencial como alimento funcional y fue mayormente aceptada por los consumidores.
Esta investigación fue financiada por la Universidad Nacional de Misiones y el Ministerio de Educación, Cultura, Ciencia y Tecnología, Argentina.

Palabras clave: Algas Marinas; Alimentos Fortificados; Proteínas; Dieta Sin Gluten; Comportamiento del Consumidor.

Entry terms: Pastas fortificadas.

KEY MESSAGES

- Seaweed powder increased nutritional values of gluten-free pasta.
- Incorporation of seaweed powder in pasta affected cooking and textural properties.
- Specific dietary fiber and protein digestibility determinations for seaweed were used.
- *Pyropia columbina* seaweed 6.0% developed best technological properties into pasta.

INTRODUCTION

Today, consumers demand foods providing basic nutritional needs, but also provide a benefit to their health.

Gluten-free (GF) foods are required by people with health problems (allergies and intolerances to wheat, celiac disease) or by tendency. Gluten proteins are responsible for the elasticity and extensibility of the dough; their absence in GF pasta results in technological and quality problems[1]: higher cooking loss (macro and micronutrients loss), texture changes and low nutritional quality.[2] GF foods are very often nutritionally unbalanced, providing excess calories and lipids with a low percentage of proteins and dietary fiber.[3]

Therefore, they should be fortified with supplements from various sources rich in protein, such as legume flours[4], whey concentrates[5], egg-proteins[1], among others, to enhance their nutritional properties. The formulation of this study was developed with regional ingredients of low nutritional and technological value, for which it was optimized through various investigations.[6,7]

In last years, various investigations have addressed the implementation of different seaweeds to achieve food products based on wheat flour with better nutritional properties.[8-10] They are a potential source of prebiotics and proteins with amino acids of nutritional interest[11], in addition, they have low content in available carbohydrates and lipids[12]; all this characteristics positions seaweed as a superfood.

Pyropia columbina, is an edible red seaweed that represents a renewable resource from the Argentine Patagonian coast; it provides a considerable protein content, has better chemical score than cereals, similar protein digestibility to others plant foods and high level of total dietary fiber, among others properties.[13]

The aim of this study was to increase the nutritional value of gluten-free pasta through the incorporation of different amounts of *Pyropia columbina* seaweed powder and to analyze the effect of the seaweed incorporation on cooking quality, textural and sensory properties, to select the best formulation.

METHODOLOGY

Ingredients

All materials used in this work were GF and regional (Argentina). Doughs were made using cassava starch (Montecarlo®) and corn flour (Indelma®), salt (Dos Anclas®), whole milk powder (Ilolay®), vegetable fat (Margadan®), whole egg and xanthan gum (Parafarm®). *Pyropia columbina* seaweed (Patagonia) was used as a nutritional source.

The sampling of the seaweed was performed as follows: *Pyropia columbina* was collected from Punta Maqueda, inside San Jorge Gulf, Argentina. The collection was carried out from August to October 2018. It was cleaned with tap water to remove remained salt and residues, then it was washed with distilled water to remove epiphytes and encrusting material. Thereafter, it was desiccated to 4.06% moisture content at 50 ºC (Dalvo, model EHRF, Argentina) in the laboratory of Universidad Nacional de la Patagonia San Juan Bosco (Chubut, Argentina). Finally, seaweed was pulverized with a miller (VivTek, China) to get powder and stored in zip bags at -20 ºC until their use in the laboratory of Posadas, Misiones.

Commercial GF pasta of similar characteristics (ribbon type, GF, made from corn flour and cassava starch 70:30 ratio) was used for sensorial consumer's analysis (Elca®).

Pasta elaboration

Pasta was made with cassava starch and corn flour 80:20 ratio, whole milk powder (7%); salt (0.5%); xanthan gum (0.8%). Vegetable fat (3.5%), whole egg (31%) and a quantity of tap water (25–38%) were added to the solid mixture, sufficient to give dough with necessary consistency for its lamination and cutting into flat strands (ribbon) with a laminator (Pluselectric®, China). A unifactorial design was selected to study the effects of addition *Pyropia columbina* powder at two levels (6.0% and 9.0%) based on the protein content of the control pasta (without seaweed powder 0%) and seaweed samples (26.00% on dry basis; N x 6.25).

Percentage of ingredients was expressed as grams ingredient/100 g cassava starch-corn flour mix.

Methodology

Cooking quality parameters (optimal cooking time (OCT), cooking loss and water absorption) for each formulation was determined through AACCI Method 66-50.01[14], by triplicate. The OCT is the limit time where the opaque line in the center of pasta disappears.

Pasta samples were immersed in boiling water to each OCT, then drained in a strainer and distilled water was added to adjust its volume to 200 mL, followed by evaporation of the cooking water at 105 ºC. The residue was weighed and represented as percentage of the original pasta sample. Water absorption of drained pasta was determined as [(cooked pasta weight – raw pasta weight) / raw pasta weight] x 100.

TA XT2i Texture Analyzer (Stable Micro Systems, UK) was used to detect changes in the textural properties of pasta. Ten repeated measurements were taken for each formulation and the data were analyzed using Texture Exponent software version 1.22 (Stable Micro Systems). The force required to shear the pasta was measured on four ribbon cooked pasta using the Light Knife Blade A/LKB probe to simulate the biting of incisive teeth (100% compression; speed=0.17 mm/s).[6] Shear force (N.s) was calculated as the area under the curve of the force (N) versus time (s) graph obtained from software. For texture parameters determination, each ribbon pasta was cut to 4 cm long and subjected to two compression cycles test equipped with compression probe (P/75), using a 50 kg load cell. The pre-test speed was 1 mm/s, while the test speed was set on 0.5 mm/s, and the strain was 75%. From the force–distance curve, firmness (N), springiness and chewiness were measured.

An analytical sensory analysis was carried out to select the best pasta formulation, also considering the quality cooking and texture parameters, to continue the study. Sensory evaluation of pasta was performed at laboratory of Facultad de Ciencias Exactas, Químicas y Naturales, Posadas, Argentina. Each pasta sample was cooked in boiling water with salt, to their OCT and 20 g were placed in thermal plastic cups. All samples were presented in a sequential monadic manner following a randomized order across participants, identified with 3-digit codes. The analytical intensity test was performed by eight semi-trained judges, who have been involved in our research for more than ten years, for descriptive evaluation of textural quality between control pasta and two pasta samples with different concentrations of seaweed powder. The sensory attributes evaluated were hardness, adhesiveness and chewiness; the definitions were adopted from Torricella Morales et al (2007).[7] Each attribute was evaluated on a 7-point scale (from very low (1) to very high (7)) on a survey form. Test was carried out in triplicate.

Cooked pasta (control and pasta with selected seaweed concentration) were evaluated using Official Methods of Analysis AOAC[15]. The moisture content (950.46 B), ash (923.03), protein (N × 6.25) content was converted from nitrogen measurements by the Kjeldahl method (984.13) and fat (922.06). Total dietary fiber was determined according to Prosky et al.[16] method modified by Lahaye[17], and control (AOAC, 991.43).[15] Carbohydrates were calculated by difference (100 - [moisture + fat + protein + dietary fiber + ash]) and caloric value was calculated based on the composition, using the Atwater conversion factors of 4.0 kcal/g for protein, 9.0 kcal/g for fat and 4.0 kcal/g for available carbohydrates.

For *in vitro* protein digestibility, control pasta was incubated with pepsin (0.1 mol equiv/L HCl, 37 ºC, 3 h) and pancreatin (0.2 mol/L phosphate buffer, 37 ºC, 24 h, pH 8) according to Akeson & Stahmann method with modifications.[18] The total protein concentration from the supernatant was studied by Kjeldahl method, on dry basis and the *in vitro* protein digestibility was calculated as $[N_{TD} / N_T]$ x100, where N_{TD}: total proteins of digested pasta and N_T: total proteins of undigested pasta.
Due to the presence of seaweed in the pasta formulation in study, the Rudloff & Lönnerdal[19] method was used to determine the *in vitro* protein digestibility (%) in these samples. It was calculated as [(NPNs - NPNis) / (Nt - NPNis)] x 100.

NPNs: non-protein nitrogen soluble in 20% of trichloroacetic acid after digestion with pepsin and pancreatin enzymes; Nt: total nitrogen of pasta, and NPNis: initial non-protein nitrogen soluble in 20% of trichloroacetic acid from pasta.
The nitrogen content (NPNs, Nt and NPNis) was determinate using Kjeldahl method (N × 6.25). Both determinations by triplicate.

The consumer acceptability between a commercial pasta and the enriched pasta was evaluated to determine the impact of the incorporation of seaweed. The test was carried out in the laboratory facilities of the same Faculty, according to the ISO/IEC 17025 standard.

Fifty-two consumers of pasta (all of them consumed pasta at least once a week; nineteen out of them, were celiac), tested the cooked pasta to their OCT. Pasta was served immediately with a three-digit code, one sample at a time following a randomized order. Consumers answered marking the option choice pasta in terms of external appearance (visual and olfactory evaluation), flavor, mouth feel (chalkiness and stickiness) and acceptability, using a 5-point hedonic scale with 5-like very much, 3- neither like nor dislike, 1-dislike very much. At the end of the form, the word "observations" was added, so that consumers could add what they consider important or strange when evaluating the pasta.

Data analysis

Data obtained of cooking and texture quality, proximate analysis and protein digestibility, were subjected to statistical analysis using one factor analysis of variance (ANOVA) and to determine the significances within treatments, Fisher's multiple range test was used (P<0.05), through Statgraphics software plus 5.1.

In sensory analysis, a score was assigned to each category of the scale (1 for the lowest and 7 for the highest in the analytical test, and 1 for the lowest and 5 for the highest in consumer acceptability test). Data were analyzed using ANOVA and Fisher's multiple range test, the results were presented as a radar chart.

RESULTS

The cooking quality parameters analyzed of control pasta and seaweed-enriched pasta are shown as mean value and standard deviations in Table 1.

Table 1 Quality characteristics of gluten-free pasta. Means and standard deviations of water absorption and cooking loss (n=3).

Pasta	OCT (min)	Water absorption (%)*	Cooking loss (%)*
SW-0	7.0	101.3±4.6[a]	3.6±0.3[a]
SW- 6	5.0	81.9±2.8[b]	2.8±0.2 [b]
SW-9	5.0	81.30±0.60[b]	2.90±0.20[b]

*Different letters in same column show significant difference among mean value ± standard deviation (P< 0.05).
OCT: Optimal Cooking Time. SW-0: control pasta without seaweed; SW-6: pasta with 6.0% seaweed; SW-9: pasta with 9.0% seaweed.

The cooking quality parameters analyzed did not present statistically significant differences between the enriched samples for water absorption and cooking loss (P>0.05), but decreased according to control pasta (Table 1).

Results obtained of textural parameters of control pasta and seaweed-enriched pasta are shown as mean value and standard deviations in Table 2.

Table 2 Quality textural characteristics of gluten-free cooked pasta expressed as the mean value and standard deviations (n=10).

Pasta	Firmness (N)	Springiness	Chewiness	Shear force (N.s)
SW-0	52.0±3.40[a]	0.70±0.04[a]	23.0±2.30[a]	19.9±1.00[a]
SW- 6	51.5±3.80[a]	0.60±0.03[b]	18.8±2.20[a]	20.4±1.30[a]
SW-9	16.4±1.30[b]	0.80±0.05[c]	9.10±0.90[b]	5.50±0.40[b]

Different letters in same column show significant difference among mean value ± standard deviation (P<0.05).
SW-0: control pasta without seaweed; *SW-6*: pasta with 6.0% seaweed; *SW-9*: pasta with 9.0% seaweed.

With 6.0% seaweed powder incorporated to the formulation (SW-6), firmness and shear force of pasta did not show significant differences (P>0.05) regarding to control; nonetheless, higher seaweed concentration (SW-9), there was a great decrease in all texture parameters, except springiness (Table 2).

Figure 1 shows the results obtained from Intensity test of the textural characteristics of control pasta and seaweed-enriched pasta at two levels:

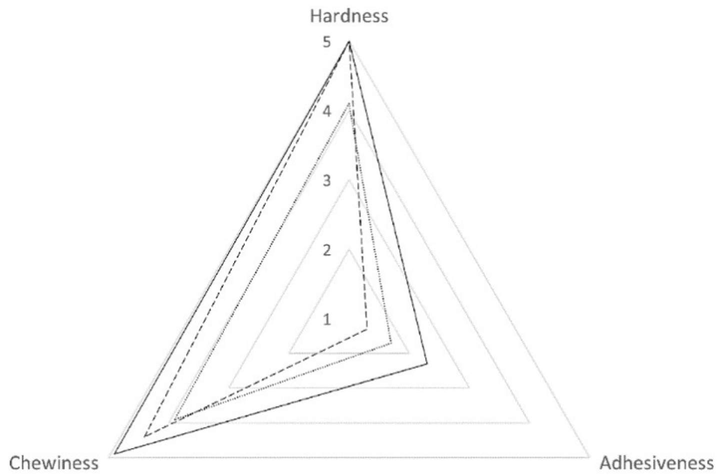

Fig 1. Results from the sensory test of intensity of texture attributes of cooked pasta.
SW-0: pasta control (continuous line); SW-6: pasta with 6.0% seaweed powder (dashed line); SW-9: pasta with 9.0% seaweed powder (dotted line).

To higher seaweed concentration, higher adhesiveness and lower hardness and chewiness for pasta were recognized in sensory analysis, with significance difference (P<0.05) regarding control. The texture attributes perception for SW-6 concurred with the desirable textural parameters (firmness, springiness, chewiness and shear force) and lower cooking loss; their results were less affected, so the following studies were performed with this formulation.

Results of the nutritional composition of formulations are presented as mean values and standard deviations in Table 3.

Table 3 Nutritional composition of control pasta (SW-0) and seaweed-enriched pasta (SW-6). Means ± standard deviation (n = 3).

Pasta	Moisture	Protein*	Ash*	Total dietary fiber*	Fat*	Available carbohydrates	Caloric value (kcal/100g)
SW-0	68.8±0.2^{a}	2.1±0.2^{a}	0.4±0.1^{a}	3,0±0.2^{a}	0.4±0.4^{a}	25.0±1.1	112,74
SW-6	70.6±0.1^{a}	2.6±0.1^{b}	1.0±0.1^{b}	4.5±0.0^{b}	0.1±0.0^{b}	21.0±0.9	95,48

* (% w.b.): percentage on wet basis.
Different letters in same column show significant difference among the values (P<0.05).
SW-0: control pasta without seaweed; SW-6: pasta with 6.0% seaweed.

Cian et al.[13] pointed out that *Pyropia columbina* is low-fat, and rich in protein, minerals and dietary fibers. Therefore, it could be expected that addition of this seaweed powder to the GF pasta analyzed favored their chemical composition in terms of the proteins, fibers and ash, with significant differences (P<0.05) between samples (Table 3); the higher ash content indicate the presence of minerals in pasta. Caloric value decreased due to the decrease in available carbohydrates but also due to the great decrease in fat in pasta.

Protein digestibility of the SW-6 was 99.3% (± 4.2); it improved with the addition of seaweed powder, in relation to the value obtained from the control pasta (92.3 ± 3.8%); an increase in mean value was obtained, but it did not present significant differences with respect to the control pasta (P>0.05).

Results of the acceptability sensorial analysis are presented in Figure 2.

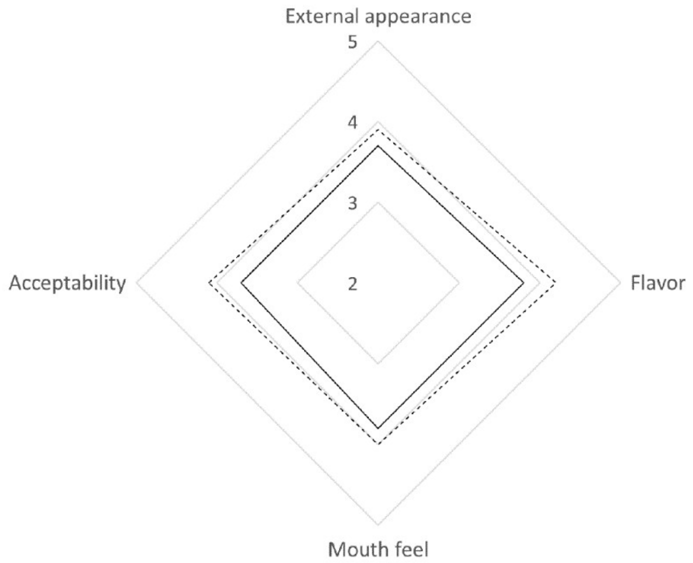

Fig.2. Results from the acceptability sensorial analysis of GF pasta with consumers. Commercial pasta (continuous line); SW-6: pasta with 6.0% seaweed powder (dashed line).

The results of the sensory evaluation test revealed that SW-6 received the highest score in flavor and acceptability. Significant differences between commercial pasta and seaweed-enriched sample were not found for neither external appearance nor mouth feel (P>0.05); panelists were not able to perceive any difference between pastas analyzed through the view, neither smell or chew (texture).

DISCUSSION

The results obtained in cooking quality, texture and sensory test served to choose the best formulation; they indicated that the ingredients formed a good network to maintain the conditions required for quality pasta.

Considering that OCT is the necessary time to obtain complete gelatinization of starch, the non-starch polysaccharides from the added seaweed to pasta is likely to result in a lower cooking time. Cooking loss is referred as the total contents of solids that leach into the cooking water; this may be due to both: amylose leaching and solubilization of some soluble proteins. Starch gelatinization occurs inward and it occurs at a rapid rate at low protein concentrations[20], however, a contrary behavior to what was expected in water absorption was observed when it was enriched with seaweed powder. The lower water absorption and cooking loss in the matrix could be due to the strong interaction of phycocolloids and proteins naturally present in seaweed[13], with xanthan gum and egg proteins in the formulation analyzed, which would form a very stable network.

Regarding to textural parameters, the higher content of seaweed fibers probably led to heterogeneity of dough, causing the difference in the analyzed textural parameters in SW-9 pasta. In enriched- pasta, a decrease in SW-6 and an increase at higher the concentration of seaweed (SW-9) was observed in the springiness parameter; this could be explained by the increase in seaweed fibers content that cause disruption in the structure of the pasta[21,9] and a incorrect reading. This higher fiber content could also be responsible for the increase in adhesiveness observed in the analytical sensory analysis in SW-9 because the fiber absorbs water and can become adhesive.[22] The desirable textural characteristics of noodles include firmness and springiness, which in fresh pasta comes mostly from the stable and flexible protein-starch matrix.[21]

The increase in protein in 1.2 times, ash in 2.5 and total dietary fiber in 1.5 times regarding to the control pasta, was as expected considering the chemical composition of seaweed used: protein 28%, total dietary fiber 48.02% and ash 6.46%, dry basis.[13] Cassava starch and corn flour used contributed with 0, 0 and 0.4 g/80g and 1.0, 0.6, 0.2 g/20g to protein, fat and dietary fiber, respectively, in the analyzed mixture. The decrease in fat content was also observed by other authors when adding seaweed powder to their noodle made with wheat flour[9,21] and those GF pasta.[22] The lower fat content in the seaweed pasta may be due to dilution by the high water retention capacity of the seaweed powder,[9,21] when dough was prepared; but it could also be due to the higher fiber content that formed a strong network that retains fat and prevents its determination.

In vitro protein digestibility of seaweed *Pyropia columbina* was 74.3% and authors attributed this value to fiber content (48%), which could block the access of digestive enzymes and decrease the activity of proteolytic enzymes[13]. This effect was not observed in the studied pasta due to milling seaweed and cooking pasta processes that could have facilitated the enzymatic attack.

Texture and appearance of pasta after cooking are the most important quality parameters for the consumers; furthermore, according to Lemes et al.[23], flavor and odor influence its acceptance. When seaweed is integrated into foodstuffs, odor and flavor are important aspects to consider, due to seaweeds are especially well-known for their umami taste, which can also enhance the intensity of others flavors. However, consumers expressed pleasure in consuming something different and enjoyable in every way.

CONCLUSIONS

Addition of *Pyropia columbina* seaweed powder at 9.0% to this pasta formulation was unsuitable from the physical, textural and analytical-sensory point of view, while the addition of seaweed powder at 6.0% developed the characteristics of quality required for pasta. In addition, the nutritional content of the pasta was significantly improved in terms of protein (by 23% increase), dietary fiber (by 50%) and ash (by 150%) regarding to the control pasta. These results show the great potential of this pasta as a functional food. Flavor was the factor that most influenced general acceptance. With the SW-6 pasta, the concept that gluten-free foods have low nutritional value is reversed.

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AUTHOR´S CONTRIBUTION

L.B-M. was responsible author and wrote the manuscript with the collaboration of M.M-E. and O.J-E.; B.R-G. and F.M-A. have contributed with bibliography search and reviewed different stages of writing.

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CONFLICTS OF INTEREST

The authors state that there are no conflicts of interest in preparing the manuscript.

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