**Food derived protein amyloid-like fibrils and their characterization**

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The increasing interest in novel, more nutritious and more sustainable food consumption leads to the pursuit of new approaches to incorporate alternative proteins into the human diet more efficiently. Food-derived protein amyloid-like fibrils (PAFs) are rod-shape fibrous aggregates that formed under acidic conditions and high temperatures in prolonged processing times (Xu et al., 2023).Among the other applications, PAFs can be incorporated into emulsions to increase the stability due to their long length, flexibility and high hydrophobicity, depending on the protein type 2. In this study, PAFs were produced from different protein sources – i.e., potato, green lentils, pea, spirulina and whey proteins – applying heating treatments at 90 ºC with a heating time of up to 10 hours. Their formation was evaluated through fluorescence microscopy, fluorescence intensity by ThT fluorescence assay, and changes in protein structure were followed through circular dichroism. The structure of PAFs were evaluated through transmission electron microscopy (TEM). Fluorescence intensity evaluation confirmed that the prolonged heating time showed increased fibril formation for whey, pea, and potato protein. Additionally, TEM analysis confirmed the formation of nanofibrils after 2h heating time. Thus, it was possible to produce protein nanofibrils using different sources of vegetable proteins. This study shows that, depending on the protein source, methodological aspects of fibril formation need to be carefully designed in accordance with the observed structural transitions.

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**References**

1. Xu, Y. *et al.* Food-derived protein amyloid-like fibrils: Fibrillation mechanism, structure, and recent advances for the stabilization of emulsions. *Food Hydrocoll* **145**, 109146 (2023).

2. Yue, J. *et al.* Recent advances of interfacial and rheological property based techno-functionality of food protein amyloid fibrils. *Food Hydrocoll* **132**, 107827 (2022).