AUCTORES
Globalize your Research

Journal of Nutrition and Food Processing

Aouatif CHENTOUF *

Open Access

Research Article

Antioxidant Activity of Food Protein: Yellow Pea Protein Isolate NUTRALYS® in a cell free Environment

Aouatif CHENTOUF[1*], Damien TRUFFIN[1], Sandrine COTIER[2], Maryse MARTIN[2] and Manilduth RAMNATH[2]

[1]*Roquette Frères, 1 rue de la Haute Loge, 62136 Lestrem, France*

[2]EUROFINS CEREP, Le Bois L'èvêque, 86600 Celle-L'Evescault, France

***Corresponding author:** Roquette Frères, 1 rue de la Haute Loge, 62136 Lestrem, France.

Received date: November 17, 2021; Accepted date: December 02, 2021; Published date: December 08, 2021

Citation: Aouatif CHENTOUF, Damien TRUFFIN, Sandrine COTIER, Maryse MARTIN and Manilduth RAMNATH (2021). Antioxidant Activity of Food Protein: Yellow Pea Protein Isolate NUTRALYS® in a cell free Environment. *J. Nutrition and Food Processing*, 4(8); DOI:10.31579/2637-8914/075

Abstract:

In recent years, the demand for "all natural" products is increasing because of the increasing limitations on the use of synthetic antioxidants and enhanced public awareness of health issues. Many natural food components like oils and oilseeds, proteins and protein hydrolysates, fruits and vegetables, oat and rice bran, spices, herbs and tea have antioxidant properties. Natural antioxidants from these food components provide oxidative stability to the food product. The antioxidant activity of proteins is mainly due to interactions between their ability to inactivate reactive oxygen species, chelate pro-oxidative transition metals, scavenge free radicals, and reduction of hydroperoxides. The objective of this study was to verify the antioxidant potential of Pea Protein Isolate 'NUTRALYS® S85F' through different *in vitro* tests:

- TOTAL ANTIOXIDANT RADICAL SCAVENGING ACTIVITY (ORAC ASSAY);
- SUPEROXIDE RADICAL SCAVENGING ACTIVITY (NBT ASSAY);
- LIPID PEROXIDATION INHIBITION ASSAY (MDA KIT);
- HYDROXYL RADICAL SCAVENGING ACTIVITY (HRS ASSAY);
- NITRIC OXIDE SCAVENGING ABILITY

NUTRALYS® S85F clearly demonstrates intrinsic antioxidant activities as can be observed from its scavenging activity toward the peroxyl radicals (ORAC), scavenger of the superoxide radicals (NBT), hydroxyl radical scavenging (HRS), prevention of lipid oxidation (MDA) and nitric oxide scavenging (NO) capabilities. Based on our results, NUTRALYS® S85F will provide oxidative stability to food products and health benefits to the consumer.

Key words: pea protein isolate; antioxidant activities; natural component; nutralys ®s85f; orac test; nbt assay; lipid peroxidation inhibition assay; hydroxyl radical scavenging; oxidative stability

Introduction

A free radical can be defined as any molecular species capable of independent existence that contains an unpaired electron in an atomic orbital. Many radicals are unstable and highly reactive. The most important oxygen-containing free radicals in many disease states are hydroxyl radical, superoxide anion radical, hydrogen peroxide, oxygen singlet, hypochlorite, nitric oxide radical, and peroxynitrite radical. These are highly reactive species, capable in the nucleus, and in the membranes of cells of damaging biologically relevant molecules such as DNA, proteins, carbohydrates, and lipids. A role of oxidative stress has been postulated in many conditions, including anthersclerosis, inflammatory condition, certain cancers, and the process of aging. Oxidative stress is now thought to make a significant contribution to all inflammatory diseases (arthritis, vasculitis, glomerulonephritis, lupus erythematous, adult respiratory diseases syndrome), ischemic diseases (heart diseases, stroke, intestinal ischemia), hemochromatosis, acquired immunodeficiency syndrome, emphysema, organ transplantation, gastric ulcers, hypertension and preeclampsia, neurological disorder (Alzheimer's disease, Parkinson's disease, muscular dystrophy), alcoholism, smoking-related diseases, and many others. The human body is in constant battle to control free radicals steady state

In recent years, the demand for "all natural" products is increasing because of the increasing limitations on the use of synthetic antioxidants and enhanced public awareness of health issues. Many natural food components like oils and oilseeds, proteins and protein hydrolysates, fruits and vegetables, oat and rice bran, spices, herbs, and tea have

antioxidant properties. Natural antioxidants from these food components provide oxidative stability to the food product. The antioxidant activity of proteins is mainly due to interactions between their ability to inactivate reactive oxygen species, chelate pro-oxidative transition metals, scavenge free radicals, and reduction of hydroperoxides.

The objective of this study was to verify the antioxidant potential of Pea Protein Isolate 'NUTRALYS®' through different *in vitro* tests.

Materials and Methods

NUTRALYS® was weighed and reconstituted at a final concentration of 10 mg/ml in 100 % H_2O (stock solution) and mixed for 30 minutes at room temperature, this was followed by centrifugation and the clarified supernatant was diluted and used for the subsequent assays:1, 10, 20, 100, 200, 500, 1,000 µg/ml final concentration for all assays. The above-mentioned stock solutions were separated into several aliquots and stored at - 20 °C. Once an aliquot was thawed it was immediately used for a specific assay and never re-frozen.

All dilutions prepared for each experiment were maintained at room temperature until use in the assay.

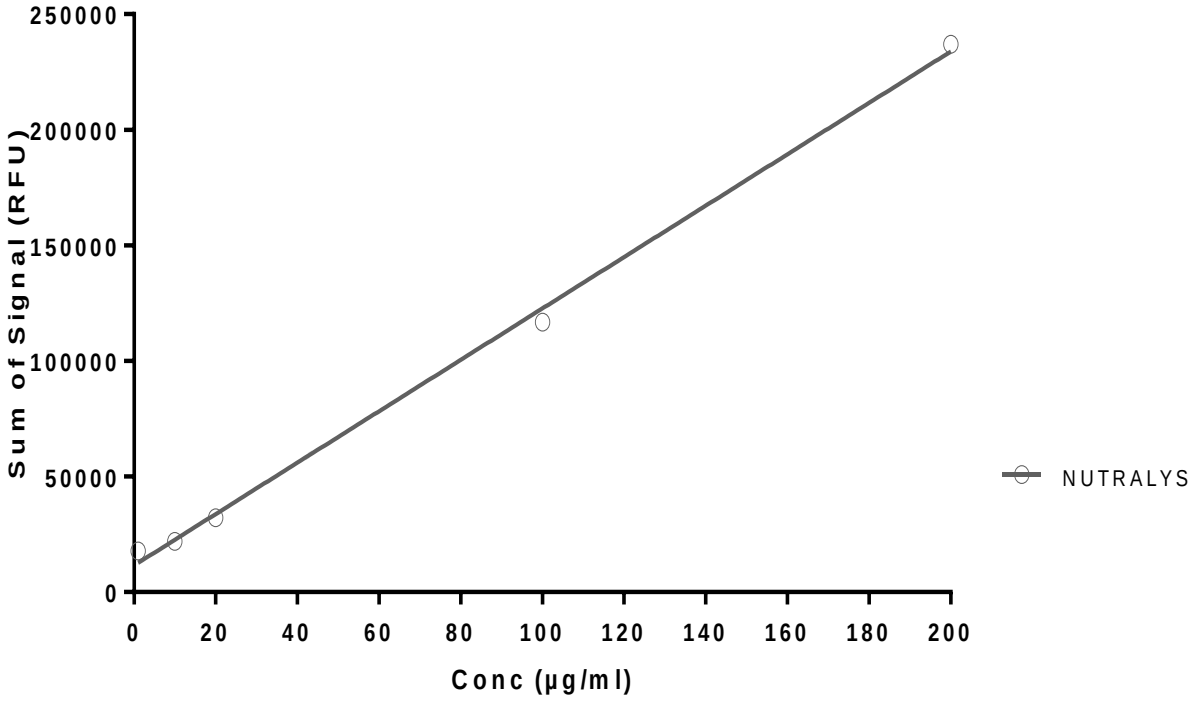
Catechin, Ascorbic acid, Quercetin and Trolox are used as positive controls.

Results & Discussion

Total antioxidant radical scavenging activity (ORAC assay):

Antioxidant activity of NUTRALYS® was determined by the ORAC method, based on BMG Labtech protocol. The method measures the antioxidant scavenging activity against the peroxyl radical, induced by 2, 2'-azobis-(2-amidinopropane) dihydrochloride (AAPH), at 37°C for 30 minutes.

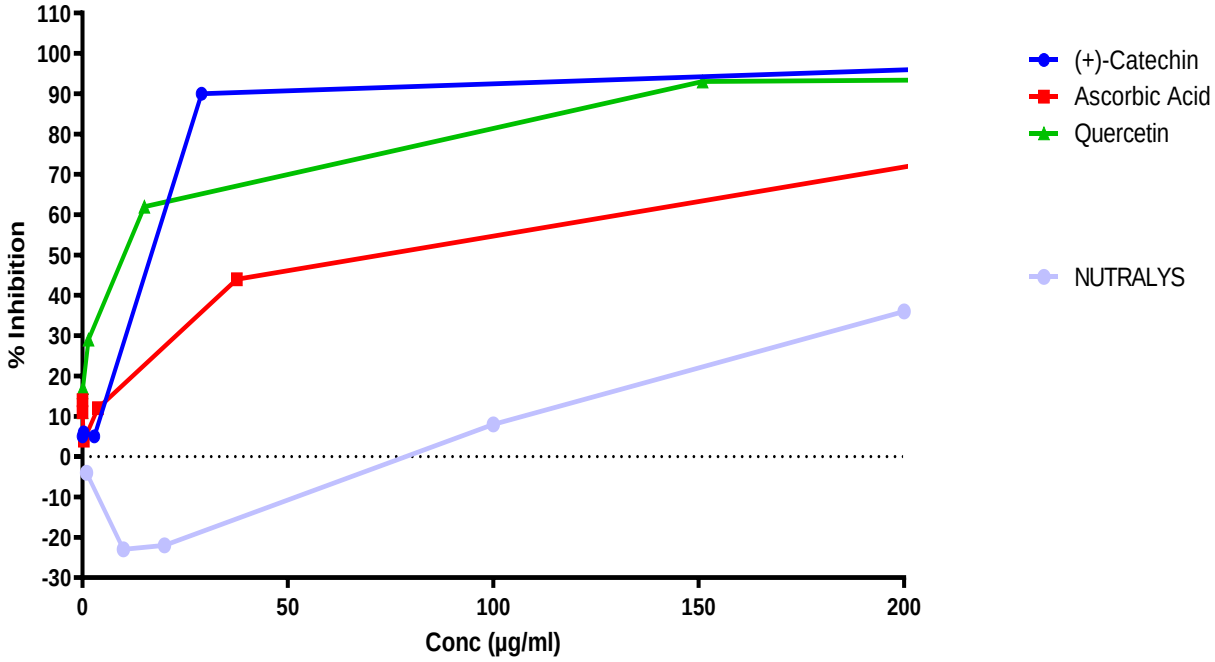
TE over considered concentration range = Slope regression curve (Trolox®) / Slope regression curve (sample)



NUTRALYS® has significant scavenging activity toward the peroxyl radicals however, the Trolox equivalent is significantly lower than that of any of the reference antioxidants

Lipid peroxidation inhibition assay (MDA kit):

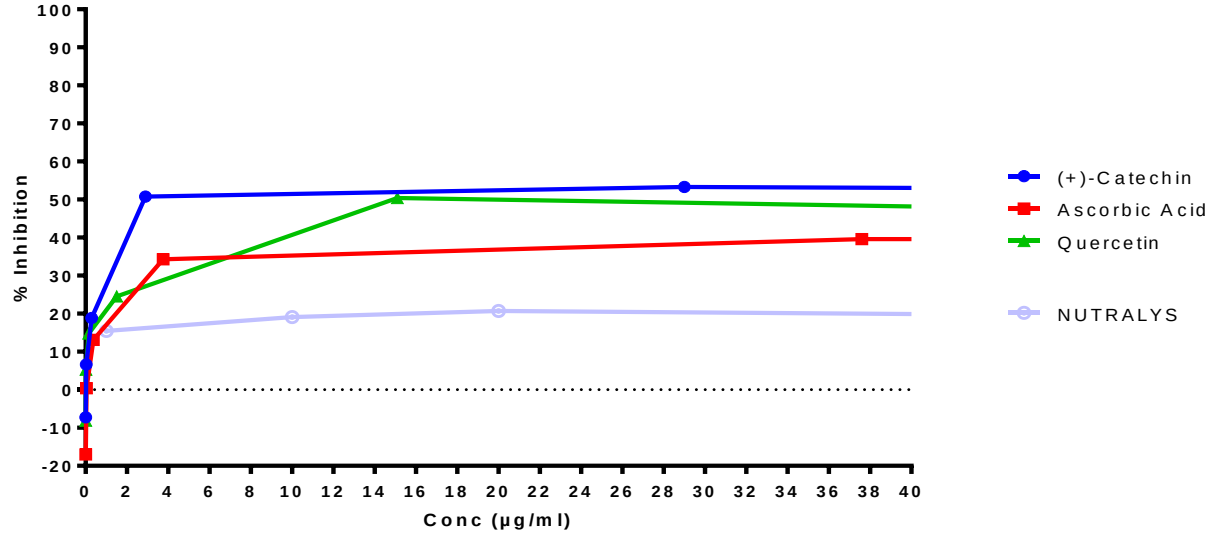
The lipid peroxidation inhibition assessment of NUTRALYS® was performed using the MDA assay kit (Sigma/MAK085)



NUTRALYS® has a moderate effect on the prevention of lipid oxidation when compared to antioxidant references

Nitric oxide scavenging ability:

Nitric oxide scavenging activity can be estimated by the use of Griess IlIosvoy reaction (Garrat, 1964) as described by Parul et al. (2013),



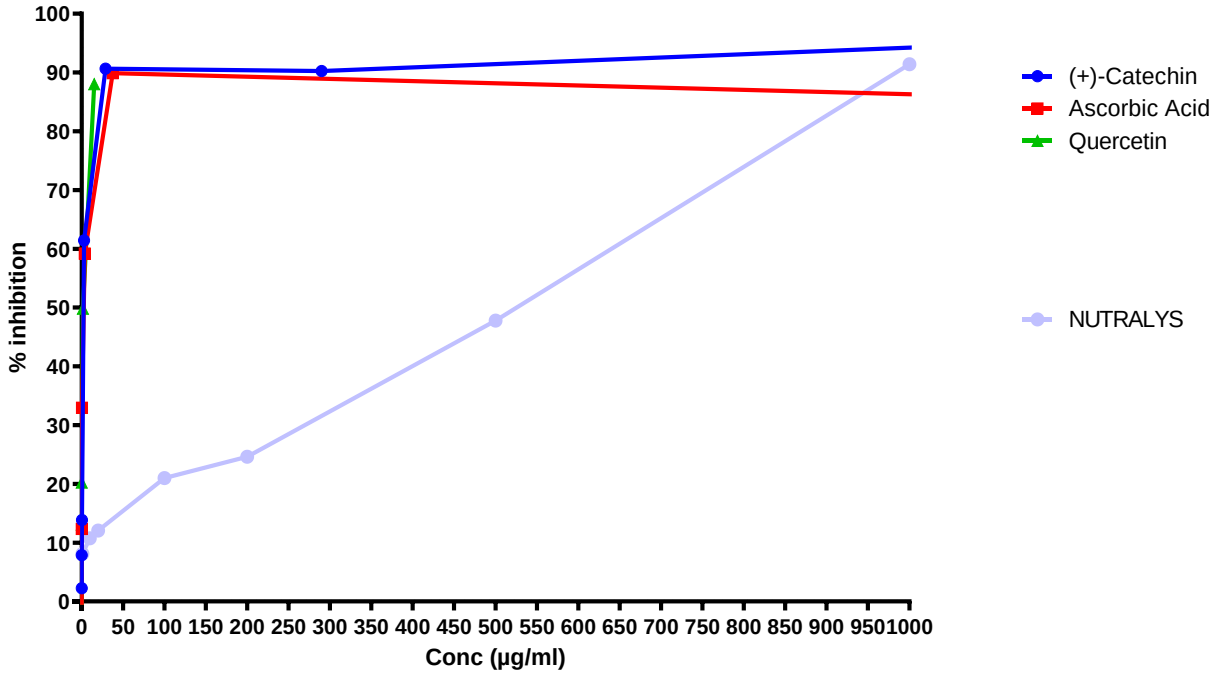
NUTRALYS® does demonstrate scavenging of the NO when compared to antioxidant references

Superoxide radical scavenging activity (NBT assay):

The superoxide radical-scavenging activity of NUTRALYS® was determined according to the method of Chang et al. (2001) and Tung et al. (2009),

The percent inhibition was calculated according to the following equation:

% inhibition = [(absorbance of control - absorbance of sample) / absorbance of control] x 100.

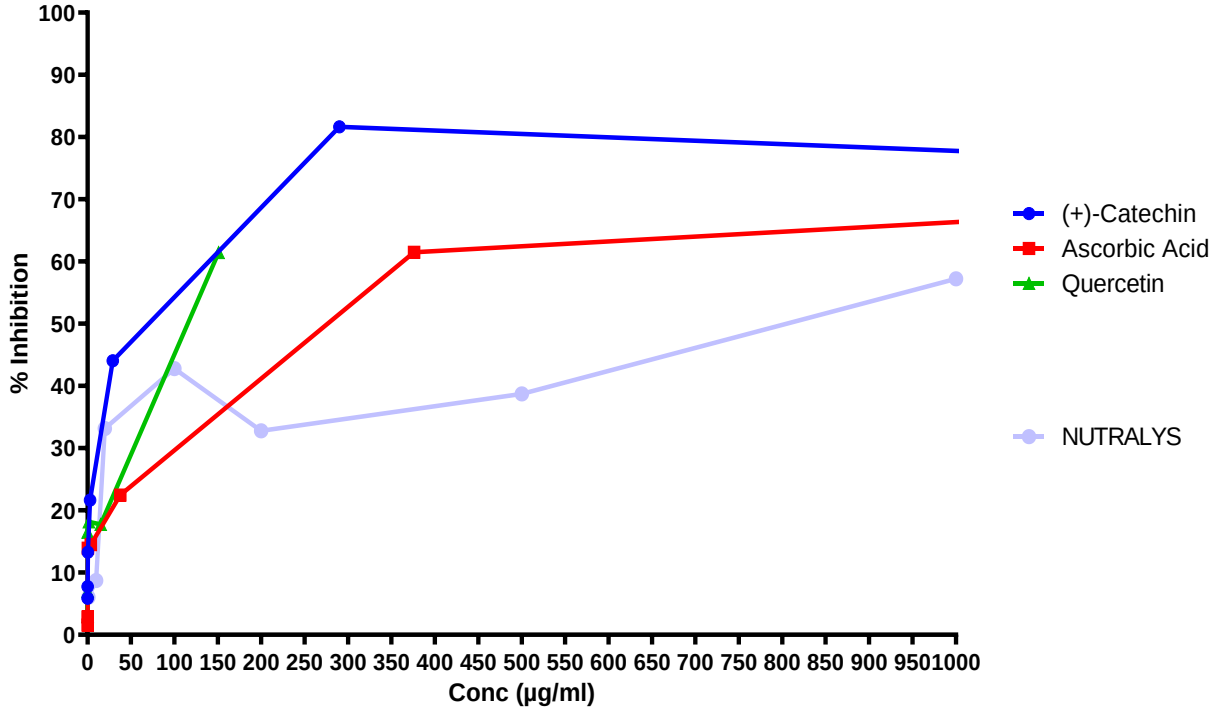


NUTRALYS® at the highest test concentration (1 mg/ml) displayed 90% inhibition.

Hydroxyl radical scavenging activity (hrs assay):

NUTRALYS® was tested at 7 concentrations in presence of iron-EDTA solution (mix solution: ferrous ammonium sulfate/EDTA/DMSO/ascorbic acid), as described by Halliwell and Gutteridge (1981)

NUTRALYS® has an equivalent antioxidant effects toward this particular ROS when compared to antioxidant the references



Results:

NUTRALYS® has an equivalent antioxidant effects toward this particular ROS when compared to antioxidant the references

% Inhibition	ORAC	NBT	HRS	MDA	NO
NUTRALYS® S85F	0.01*	91% at 1000 µg/ml	68% at 1000 µg/ml	39% at 1000 µg/ml	21% at 20 µg/ml

- ✓ Significant scavenging activity toward the peroxyl radicals (ORAC)
- ✓ A scavenger of the superoxide radicals (NBT) , with 90% inhibition at the highest test concentration (1000 µg/ml)
- ✓ Significant hydroxyl radical scavenging (HRS) activity however, higher concentrations of test compound was required to produce the same antioxidant effect as the reference compounds.

Conclusion

NUTRALYS® clearly demonstrates intrinsic antioxidant activities as can be observed from its scavenging activity toward the peroxyl radicals (ORAC), scavenger of the superoxide radicals (NBT), hydroxyl radical scavenging (HRS), prevention of lipid oxidation (MDA) and nitric oxide scavenging (NO) capabilities. Based on our results NUTRALYS® will provide oxidative stability to food products and health benefits to the consumer.

References

1. **H**alliwell B, Gutteridge JM. Formation of thiobarbituric-acid-reactive substance from deoxyribose in the presence of iron salts: the role of superoxide and hydroxyl radicals. FEBS Lett. 1981 Jun 15; 128(2):347-352.
2. Garrat DC. (1964) The Quantitative analysis of Drugs. Chapman and Hall Ltd., Japan, 3: 456 - 458
3. **R**ozina Parul, Sukalayan Kumar Kundu, Pijush Saha. In Vitro Nitric Oxide Scavenging Activity Of Methanol Extracts Of Three Bangladeshi Medicinal Plants. Pharma Innovation 2013; 1(12):83-88.
4. **S**hang-Tzen Chang, Jyh-Horng Wu, Sheng-Yang Wang, Pei-Ling Kang, Ning-Sun Yang, and Lie-Fen Shyur. Antioxidant Activity of Extracts from Acacia confusa Bark and Heartwood. Journal of Agricultural and Food Chemistry 2001 49 (7), 3420-3424
5. **G**utteridge, J. M., Rowley, D. A., & Halliwell, B. (1981). Superoxide-dependent formation of hydroxyl radicals in the presence of iron salts. Detection of 'free' iron in biological systems by using bleomycin-dependent degradation of DNA. The Biochemical journal, 199(1), 263–265.
6. **Y**u-Tang Tung, Jyh-Horng Wu, Ching-Yu Hsieh, Ping-Sheng Chen, Shang-Tzen Chang, Free radical-scavenging phytochemicals of hot water extracts of Acacia confusa leaves detected by an on-line screening method, Food Chemistry, Volume 115, Issue 3, 2009, Pages 1019-1024, ISSN 0308-8146

DOI:10.31579/2637-8914/075