



Proceeding Paper

Blackcurrant (*Ribes nigrum* L.) and Raspberry (*Rubus idaeus* L.) Ethanolic Extracts: Inhibitory Effects on Pancreatic Lipase and Antioxidant Activity [†]

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[†] Presented at the 4th International Electronic Conference on Foods, 15–30 October 2023; Available online:

<https://foods2023.sciforum.net/>.

Abstract: Blackcurrant (*Ribes nigrum* L.) and raspberry (*Rubus idaeus* L.) fruits, rich sources of phytochemicals such as polyphenols, have been shown to possess promising biological properties. Herein, both fruits from southern Italy have been extracted by maceration by using ethanol as the solvent and investigated for their potential pancreatic lipase inhibitory activity. The radical scavenging effects were also analysed by using the 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) tests. Our study showed that the extracts of both blackcurrant and raspberry have significant inhibitory activity against lipase with IC₅₀ of 5.1 and 30.2 µg/mL, respectively, better than the positive control orlistat (IC₅₀ of 37.1 µg/mL).

Keywords: Berries; polyphenols; bioactivity; functional food



Citation: Tundis, R.; Cappello, A.R.; Loizzo, M.R. Blackcurrant (*Ribes nigrum* L.) and Raspberry (*Rubus idaeus* L.) Ethanolic Extracts: Inhibitory Effects on Pancreatic Lipase and Antioxidant Activity. *Biol. Life Sci. Forum* **2023**, *26*, 20. <https://doi.org/10.3390/Foods2023-15065>

Academic Editor: Antonello Santini

Published: 14 October 2023



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1. Introduction

In recent years, several studies analyzing the composition, the bioactivity of red fruits, and the relationship between fruit intake and reduced risk of several chronic diseases, including obesity, have been published. Fruits are sources of not only vitamins, minerals, and dietary fiber but also several healthy phytochemicals. Blackcurrant (*Ribes nigrum* L.) and raspberry (*Rubus idaeus* L.) fruits are rich sources of polyphenols that have been shown to possess antioxidant, anti-inflammatory, antimicrobial, and hypoglycaemic properties [1,2]. In this study, in order to investigate how environmental factors may influence types and contents of active substances and to prospect a potential new use as functional foods and/or nutraceuticals, we investigated the radical scavenging effects and lipase inhibitory activity of the ethanolic extract of blackcurrant (*Ribes nigrum* L.) and raspberry (*Rubus idaeus* L.) fruits collected in southern Italy. Several previous studies have demonstrated that plants that grow in various environments produce different active substance contents because of their wide distribution in different geological zones. This will result in variations in their internal qualities, as well as in the same species from different growing regions [3].

2. Materials and Methods

Plant materials and extraction procedure. Ripe fruits were collected in southern Italy (WGS84: 39°87'13" N, 16°06'53" E). Fresh fruits (350 g) were exhaustively extracted by maceration using ethanol as a solvent (4 × 1.2 L). Dry extracts were stored in brown glass bottles and kept at 4 °C before analysis.

Total polyphenols content (TPC). The TPC was determined using the Folin–Ciocalteu method in which extracts (concentration of 1.5 mg/mL) were mixed with water, sodium carbonate 15% (*w/v*), and Folin–Ciocalteu reagent [4]. After an incubation of 2 h at room

temperature, the absorbance was read at 765 nm using a UV-vis Jenway 6003 spectrophotometer (Milan, Italy).

Total flavonoids content (TFC). For the TFC determination, extracts (concentration of 1.5 mg/mL) were added to water and sodium nitrite 5% (*w/v*). After 5 min, aluminium chloride 10% (*w/v*) was added. After another 6 min, sodium hydroxide 1 M and water were added. Then, the absorbance was read at 510 nm [4].

Radical scavenging activity. The radical scavenging activity was investigated using the 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) tests [4]. In the ABTS test, an ABTS radical cation solution was prepared and mixed with a solution of potassium persulfate and left before use for 12 h in the dark. The ABTS solution was diluted with ethanol to an absorbance of 0.70 at 734 nm. After the addition of the lavender extract to the ABTS solution, the absorbance was read after 6 min at 734 nm. In the DPPH test, a mixture of the DPPH ethanol solution (1.0×10^{-4} M) and extracts at different concentrations were prepared and kept for 30 min in the dark. The bleaching of the DPPH was determined by reading the absorbance at 517 nm. Ascorbic acid was used as positive control in both assays.

Pancreatic lipase inhibitory activity test. To investigate the lipase inhibitory activity, a previously reported protocol was adopted [4]. Concisely, extracts at different concentrations were mixed with the enzyme, Tris-HCl buffer (pH 8.5), and 4-nitrophenyl octanoate, and the mixture was incubated for 30 min at 37 °C. Then, the absorbance was measured at 405 nm. Orlistat was used as a positive control.

Statistical analysis. Experiments were performed in triplicate. The Prism GraphPad Prism Software (San Diego, CA, USA) was used to calculate the concentration causing 50% inhibition (IC_{50}). Data were analyzed using the one-way analysis of variance (ANOVA) and significant differences were calculated using Tukey's test.

3. Results and Discussion

The fresh fruits of *R. nigrum* and *R. idaeus* from Calabria (southern Italy) have been subjected to maceration by using ethanol as the solvent for obtaining extraction. Yields of 11.8 and 11.2% for the blackcurrant and raspberry, respectively, were observed. A TPC value of 501.1 and 483.7 mg chlorogenic acid equivalents/100 g plant materials for raspberry and blackcurrant, respectively, were determined. The blackcurrant extract was characterized by the highest TFC with a value of 35.1 mg quercetin equivalents/100 g plant materials compared to the raspberry (26.3 mg quercetin equivalents/100 g plant materials).

The raspberry extract was the most active in the ABTS test with an IC_{50} of 1.6 µg/mL, a value comparable to that of the positive control (Table 1).

Table 1. Radical scavenging activity (IC_{50} , µg/mL) of *R. nigrum* and *R. idaeus* extracts.

Sample	DPPH Test	ABTS Test
<i>R. nigrum</i>	3.3 ± 0.5^b	4.7 ± 0.6^a
<i>R. idaeus</i>	1.6 ± 0.1^a	8.9 ± 0.8^b
Ascorbic acid	1.1 ± 0.4	5.2 ± 0.2

Data are expressed as media $\pm$ standard deviation ($n = 3$). Differences within and between groups were evaluated by One-way (ANOVA) followed by Tukey's multiple range test. Results followed by different letters in a same column are significantly different at $p < 0.01$.

In recent years, inhibitors of pancreatic lipase have received great attention from researchers, and inhibitors from natural sources have still attracted much attention due to their wide range of sources, structural diversity, and low toxicity and side effects. The results of our study showed that the ethanolic extracts of both blackcurrant and raspberry have significant inhibitory activity against this enzyme, better than the positive control orlistat. In fact, the *R. idaeus* exhibited an IC_{50} value of 5.1 µg/mL. An IC_{50} of 30.2 mg/mL was found for the blackcurrant (Table 2). Both the extracts were more active than the orlistat (IC_{50} of 37.1 µg/mL).

Table 2. Lipase inhibitory activity (IC₅₀, µg/mL) of *R. nigrum* and *R. idaeus* extracts.

Sample	Pancreatic Lipase Inhibitory Activity Test
<i>R. nigrum</i>	30.2 ± 1.8 ^b
<i>R. idaeus</i>	5.1 ± 0.9 ^a
Orlistat	37.1 ± 1.1

Data are expressed as media ± standard deviation ($n = 3$). Differences within and between groups were evaluated by One-way (ANOVA) followed by Tukey's multiple range test. Results followed by different letters in a same column are significantly different at $p < 0.01$.

4. Conclusions

The present research points to the potential value of blackcurrant and raspberry extracts in inhibiting lipase and exerting antioxidant effects. The most promising results were obtained with the raspberry extract and will contribute toward the development of new functional foods with anti-obesity effects.

This report provides some basic evidence for the effects of ethanolic extracts of these fruits and suggests future in vivo studies for the identification of the molecules that exhibit antioxidant and anti-lipase effects and for the development of new products with beneficial health properties for the prevention and/or treatment of metabolic disorders.

Author Contributions: Methodology: R.T.; formal analysis: A.R.C., M.R.L. and R.T.; data curation: A.R.C., M.R.L. and R.T.; writing-original draft preparation: R.T.; writing-review and editing, A.R.C., M.R.L. and R.T. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Calabria Region (Italy) through the project titled “Aumento della produttività e della qualità di colture fuori suolo (CENTOZAF)” (CUP—J16G20000480005) within the framework PSR Calabria 14/20.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The authors wish to thank Azienda Agricola Grisolia and Azienda Agricola Armentano for supplying the *R. nigrum* and *R. idaeus* fruits.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Tao, Y.; Bao, J.; Zhu, F.; Pan, M.; Liu, Q.; Ping Wang, P. Ethnopharmacology of *Rubus idaeus* Linnaeus: A critical review on ethnobotany, processing methods, phytochemicals, pharmacology and quality control. *J. Ethnopharmacol.* **2023**, *302*, 115870. [[CrossRef](#)] [[PubMed](#)]
2. Cortez, R.E.; Gonzalez de Mejia, E. Blackcurrants (*Ribes nigrum*): A review on chemistry, processing, and health benefits. *J. Food Sci.* **2019**, *84*, 2387. [[CrossRef](#)] [[PubMed](#)]
3. Tenuta, M.C.; Deguin, B.; Loizzo, M.R.; Dugay, A.; Acquaviva, R.; Malfa, G.A.; Bonesi, M.; Bouzidi, C.; Tundis, R. Contribution of flavonoids and iridoids to the hypoglycaemic, antioxidant, and nitric oxide (NO) inhibitory activities of *Arbutus unedo* L. *Antioxidants* **2020**, *9*, 184. [[CrossRef](#)] [[PubMed](#)]
4. Tundis, R.; Conidi, C.; Loizzo, M.R.; Sicari, V.; Romeo, R.; Cassano, A. Concentration of bioactive phenolic compounds in olive mill wastewater by direct contact membrane distillation. *Molecules* **2021**, *26*, 1808. [[CrossRef](#)] [[PubMed](#)]

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