JMBFS

Journal of Microbiology, Biotechnology and Food Sciences

International peer-reviewed scientific online journal

Published by Faculty of Biotechnology and Food Sciences

FATTY ACID PROFILE OF COMMON CARP *(CYPRINUS CARPIO)* AFTER ADDITION OF ASTAXANTHIN TO THE FEED MIXTURE

Peter Herc[1], Juraj Čuboň[1], Matej Čech[1], Peter Haščík[1], Miroslav Kročko[1], Adam Hanuska[1]*

Address(es):
[1] Slovak University of Agriculture in Nitra, Faculty of Biotechnology and Food Sciences, Institute of Food Sciences, Tr. Andreja Hlinku 2, 94976 Nitra, Slovakia.

*Corresponding author: xhercp@uniag.sk

https://doi.org/10.55251/jmbfs.10581

ARTICLE INFO

Received 14. 9. 2023
Revised 31. 5. 2024
Accepted 8. 7. 2024
Published 1. 10. 2024

Regular article

OPEN ACCESS

ABSTRACT

Astaxanthin is a very strong antioxidant typically fed to rainbow trout mainly to alter the color of its' meat to red. In our study, we incorporate a relatively low concentration of astaxanthin into the diet of common carp to enhance the chemical parameters and oxidation stability of carp meat without altering the typical coloration of meat. Also, we observed the fatty acid profile of reared fish. We observed an increase in protein content in animals with astaxanthin dietary addition. It was not statistically significant, but we observed a lowering of fat content in experimental groups. Regarding lipid oxidation, we did not observe any significant effect of astaxanthin on malondialdehyde production in samples. Fatty acid profiles were determined by gas chromatography and then compared. Only significant differences were observed in stearic and arachidonic acid and in the experimental group with lower astaxanthin addition. Our results conclude that dietary astaxanthin has no adverse effect on fatty acid profile or oxidation stability during storage (at -18 °C). On the other hand, astaxanthin has the potential to enhance the protein and lower the fat content, even in low concentrations.

Keywords: *Cyprinus carpio*, astaxanthin, fatty acid, lipid oxidation

INTRODUCTION

The common carp (*Cyprinus carpio*) is considered one of the most significant freshwater fish species. It is commonly maintained in polyculture, semi-intensive systems alongside other cyprinidae species such as silver carp (*Hypophthalmichthys molitrix*), grass carp (*Ctenopharyngodon Idella*), and bighead carp (*Aristichthys nobilis*). Although common carp is native to Central Asia and Eastern Europe, it has spread to over 120 countries where it is now grown and consumed **(Chalamaiah *et al.*, 2015)**. Carp species dominate freshwater fish production, with common carp being the fourth most-produced fish in aquaculture globally **(Fraňek *et al.*, 2021)**. Because of its favorable composition of all basic nutritional components required in human nutrition, fish meat is an important representative of animal origin food. It has high levels of protein, fats, vitamins, and minerals. The presence of necessary fatty acids in fish meat is also critical. When compared to red meat, fish meat contains much less saturated fatty acids (SFA) which have no double bond in their structure. Fish meat is high in omega-3 long chain (LC) polyunsaturated fatty acids (-3 LC PUFA), which include at least two double bonds. Those fatty acids are very important in human nutrition **(Bušová *et al.*, 2020)**.

Carotenoids are tetra-terpenoid pigments produced naturally by higher plants, algae, fungus, and some bacteria; about 600 of them have been identified and described. Carotenoids are chemically divided into two groups: carotenes, which have primarily carbon and hydrogen in their structure, and xanthophylls, which are oxygenated derivatives. Carotenoids have critical roles in the biological system, including scavenging reactive oxygen species, quenching of the chlorophyll triplet state, light harvesting, and dissipation of surplus energy **(Chen *et. al.*, 2020)**. The oxygenated derivatives of xanthophyll are classified into three classes based on the presence of a functional group: the -OH group (zeaxanthin), the O group (canthaxanthin), or both -OH and O groups (astaxanthin [ASX] and lutein). ASX ($C_{40}H_{52}O_4$) is a xanthophyll carotenoid that is gaining popularity around the world **(Shah *et al.*, 2020)**. Astaxanthin possesses an unusual antioxidant activity, even more, significant than vitamin C, β-carotene, and α-tocopherol, because of this, it has received considerable attention in various research areas **(Kumar *et al.*, 2021)**. It has been approved by the US Food and Drug Administration (FDA) and the European Commission as a natural food additive for animal and fish feed, as well as a natural dye for animal and human diets **(Brendler and Williamson, 2019)**.

Carotenoids, especially astaxanthin, have been identified as the main pigments used in fish diet in order to obtain the desirable reddish color in flesh of fish species such as salmonids and trouts **(Stachowiak & Szulc, 2021).** Furthermore, literature has shown that astaxanthin has positive effects on fish growth, reproduction, survival as well as serving as a powerful antioxidant to immunostimulants in fish. Although much work has been done on optimizing astaxanthin usage and its effects in fish diet, many studies in this area of investigation were mainly focused on adult fish such as rainbow trouts and salmons **(Fan & Chen, 2007; Balendra & Singh, 2021; Zhu *et al.*, 2022).** Previously published works claimed that dietary administration of astaxanthin can directly or indirectly confer antioxidant activity and enhance cell-mediated and humoral immune response. Based on these reports it is suggested that astaxanthin has a potential value for aquaculture both in terms of increased growth performance, immune response, and protection against specific bacterial disease **(Kojima *et al.*, 2002; Park *et al.*, 2011; Jagruthi *et al.*, 2014).** In other study the supplementation of astaxanthin significantly improved the body redness, the skin astaxanthin concentration and the hepatic antioxidation property of discus fish **(Song *et al.*, 2016).**

In our study we incorporated astaxanthin into common carp fish diet in pellet form in the later stages of it rearing (6 months before slaughter). Astaxanthin is commonly fed to rainbow trout to color its' meat red, to resemble the meat of salmon. In our study, we decided to use a relatively low concentration to avoid changes of typical carp meat color. Our aim was to prolong the storage period of frozen common carp by retardation of lipid oxidation without changing the typical color of carp fish meat. Various effects of astaxanthin on chemical composition of common carp was also a major point of interest of our work.

MATERIAL AND METHODS

Common carp diet and slaughter

In our study 30 common carp individuals were used. All animals were reared for 12 months at fish farm in Ružomberok (Slovakia) using same feeding mixture. Our feeding mixture was prepared by company commercially preparing fish feeding mixture. After the first period, animals were divided into three groups: Carp_0 – control group without dietary astaxanthin; Carp_10 – experimental group with 10 $mg.kg^{-1}$ addition of dietary astaxanthin; Carp_15 – experimental group with 15 $mg.kg^{-1}$ addition of dietary astaxanthin. All groups were fed mixture according to their group for another 6 months. Ater 18 months, 5 animals from each group with similar weight were selected, slaughtered, and transported to Slovak university of agriculture (SUA) in Nitra. Animals were then dissected, skinned, and stored (at -18 °C) and analyzed at Institute of food science of SUA.

All animals were handled following the national legislation on animal welfare (DL n. 126, 07/07/2011, EC Directive 2008/119/EC). Common carps were slaughtered in compliance with Regulation 1099/2009 of the European Union on the protection of animals at the time of killing.

Table 1 Common carp feeding mixture composition

Feed component	%
Crude protein	15.50
Crude fat	3.70
Crude fiber	6.00
Crude ash	5.60
Lysine	0.85
Methionine	0.48
Calcium	1.00
Phosphorus	0.65
Sodium	1.26

Additional components of mixture were: Vitamin A 12000.00 IU. kg^{-1}, Vitamin D3 2000.00 IU. kg^{-1}, Vitamin E 60.00 mg. kg^{-1}, Copper (II) sulfate 4.00 mg. kg^{-1}, Manganese (II) oxide 40.00 mg. kg^{-1}, Zinc sulfate 159.00 mg. kg^{-1}, Sodium selenide 0.30 mg. kg^{-1}. Ater 12 months, astaxanthin was added to this mixture according to individual groups.

Chemical composition determination

The samples of fish meat were analyzed for basic chemical composition (water, crude protein, fat, and cholesterol content). To conduct the analysis itself INFRATEC 1265 device (Germany) was used, as suggested by **Trembecká *et al.* (2016)**. Results are expressed as g.100 g^{-1} of muscle dry matter.

TBARs assay

To determine effect of dietary astaxanthin addition on lipid oxidation of fish fillets TBARs method was used as used by authors **Jurčaga *et al.* (2022).** Results were calculated and expressed as mg of malondialdehyde (MDA) in 1kg of fish meat. TBARs assay was performed on first day of fish slaughter, then monthly for the rest of the storage period.

Fatty acid profile determination

The muscle fillets were separated from fish and used for the determination of the fatty acid (FA) composition. The fatty acid compositions of all fish muscles were determined by a direct method for fatty acid methyl ester (FAME) synthesis. The FA composition of the FAME was determined using a Gas Chromatograph (Agilent, 7890A series, USA) equipped with a flame ionization detector and a chiral capillary column (J&W Scientific, USA) as suggested by **Polákova *et al.* (2023)**. Results are expressed as g.100 g^{-1} of muscle dry matter.

Statistical analysis

To perform statistical analysis, XLSTAT software was used **(Data Analysis and Statistical Solution for Microsoft Excel, Addinsoft, Paris, France, 2019)**. Comparation of all obtained results from the individual analyses, analysis of variance (ANOVA) analysis with Duncan test was used. The level of significance α was set to 0.05 for all performed statistical tests.

RESULTS AND DISCUSSION

Chemical composition of common carp meat

Determination of chemical composition of common carp meat was conducted one day after the slaughter and dissection of fish. Observed parameters were water, protein, fat and cholesterol. We obtain very similar results in content of water groups. We observed lower content of fat in both experimental groups compared to control group. However, this difference was evaluated as not statistically significant ANOVA Duncan test with α=0.05. Similar situation was observed with cholesterol content where experimental group reached lower level of cholesterol, but not significantly lower. Only component with statistically difference was protein content. Experimental groups (both carp_10 and carp_15) reached significantly ($p \leq 0.05$) higher protein content than control group without dietary astaxanthin addition (Carp_0). Similar results were observed by author **Abdulrahman (2020)**. In his study author reported significantly higher protein content in common carp with astaxanthin enhanced feed without noticeable changes in water content. In contrast to his findings, we observed lowered fat content in experimental groups, where he reported increase. Our results are comparable to findings of **Ljubojević *et al.* (2016)** who also evaluated chemical composition of common carp fillets. Chemical composition of samples from our study is listed in Table 2.

Table 2 Chemical composition of common carp meat (g.100 g^{-1})

Group	Water	Protein	Fat	Cholesterol
Carp_0	69.39 ± 0.78^a	20.90 ± 0.49^b	4.55 ± 1.76^a	0.82 ± 0.21^a
Carp_10	70.09 ± 0.19^a	21.66 ± 0.23^a	2.77 ± 0.17^a	0.61 ± 0.64^a
Carp_15	70.97 ± 0.12^a	21.69 ± 0.18^a	2.77 ± 0.19^a	0.63 ± 0.03^a
P-value	0.20	0.03	0.10	0.13

Note: Means in a column with a different superscript letter (a,b) are statistically different (ANOVA with Duncan test, $p<0.05$)

Fatty acid profile

Determination of fatty acid (FA) profile of common carp meat was conducted one day after the slaughter and dissection of fish. We observed no significant differences among control and experimental groups in the majority of individual fatty acid detected during our analysis. Only exceptions were stearic and arachidonic acids. In both cases, meat from common carp with 10 mg.kg^{-1} dietary astaxanthin addition (Carp_10) reached lower average value than control (Carp_0) and second experimental group (Carp_15). MUFAs formed the majority of fatty acids in all observed carp groups, followed by SFAs and PUFAs. The most prevalent fatty acid in our common carp samples was oleic acid, followed by palmitic acid. This is in line with fatty acid profiles reported by other authors **(Fajmonová *et al.*, 2003; Yeganeh *et al.*, 2012).** Small variability in concentration of various fatty acid was explained by authors **Mráz and Picková (2011)**.

Table 3 Fatty acid profile of common carp meat (g.100 g^{-1})

Fatty Acid	Carp_0	Carp_10	Carp_15	P-value
Lauric (C12:0)	0.15 ± 0.01^a	0.13 ± 0.01^a	0.15 ± 0.00^a	0.07
Myristic (C14:0)	1.33 ± 0.01^a	1.33 ± 0.02^a	1.34 ± 0.02^a	0.69
Palmitic (C16:0)	24.40 ± 0.02^a	24.38 ± 0.21^a	24.49 ± 0.13^a	0.61
Heptadecanoic (C17:0)	0.26 ± 0.01^a	0.25 ± 0.03^a	0.26 ± 0.01^a	0.41
Stearic (C18:0)	11.04 ± 0.10^a	10.76 ± 0.12^b	11.02 ± 0.13^a	0.03
Oleic (C18:1 cis)	38.43 ± 3.25^a	44.45 ± 9.25^a	39.42 ± 8.25^a	0.10
Vaccenic (C18:1 trans-11)	4.65 ± 0.04^a	4.65 ± 0.06^a	4.68 ± 0.03^a	0.60
Linoleic (C18:2 cis)	5.19 ± 0.26^a	4.42 ± 1.03^a	5.08 ± 0.10^a	0.30
Conjugated Linoleic (C18:2 n-6)	0.12 ± 0.00^a	0.11 ± 0.02^a	0.12 ± 0.00^a	0.25
α-Linolenic (C18:3 n-3)	0.08 ± 0.03^a	0.09 ± 0.02^a	0.07 ± 0.03^a	0.69
Eicosenoic (C20:1 n-9)	0.09 ± 0.06^a	0.28 ± 0.17^a	0.12 ± 0.07^a	0.15
Arachidonic (C20:4 n-6)	1.52 ± 0.17^a	1.17 ± 0.18^b	1.55 ± 0.13^a	0.03
Eicosapentaenoic (C20:5 n-3)	0.07 ± 0.00^a	0.06 ± 0.01^a	0.07 ± 0.00^a	0.34
Docosapentaenoic (C22:5 n-3)	0.12 ± 0.01^a	0.13 ± 0.01^a	0.12 ± 0.01^a	0.64
Docosahexaenoic (C22:6 n-3)	0.03 ± 0.00^a	0.04 ± 0.01^a	0.03 ± 0.00^a	0.68
Omega 3	0.52 ± 0.03^a	0.57 ± 0.07^a	0.53 ± 0.04^a	0.43
Omega 6	7.34 ± 0.58^a	6.47 ± 1.12^a	7.40 ± 0.60^a	0.33
∑ SFA	34.68 ± 0.85^a	33.14 ± 1.78^a	34.89 ± 0.83^a	0.22
∑ MUFA	50.99 ± 0.59^a	53.34 ± 3.12^a	50.38 ± 0.70^a	0.18
∑ PUFA	9.99 ± 0.58^a	9.37 ± 0.74^a	10.11 ± 0.54^a	0.34

Note: Means in a column with a different superscript letter (a,b) are statistically different (ANOVA with Duncan test, $p<0.05$)

According to the authors, fish fillets are very diverse and made up of a variety of tissues, including white muscle, red muscle, adipose tissue, and skin. The lipid concentration of the tissues varies substantially, therefore the lipids are not evenly distributed throughout the fillet. In common carp, there is a significant differential in lipid concentration between white dorsal muscle (~1-2%), red muscle (16-17%), and the abdominal wall (~30%), as reflected in the FAs composition. Authors **Choubert *et al.* (2006)** in their work observed dietary addition of astaxanthin to rainbow trout *(Oncorhynchus mykiss)*. Similarly to our work, authors did not report significant difference between control an experimental group. Complex fatty acid profiles of observed common carp fish groups are listed in Table 3.

TBARs assay results

One of our aims was to observe potentially extended shelf life of common carp fish after astaxanthin dietary addition. Common carp fish meat was stored at -18 °C for 4 months. Measurement itself was conducted at 1st day and then monthly for the whole storage period. During our observation period, we did not measure significant ($p \geq 0.05$) difference among all carp fish group. Based on our result we can conclude that neither 10 or 15 $mg.kg^{-1}$ dietary addition of astaxanthin positively or negatively affect oxidation stability of frozen common carp meat. Authors **Aracati *et al.* (2022)** observed oxidation stability of fillets obtained from tilapia reared with astaxanthin enhanced feed. Authors did not observe significant differences in TBARs values of control and both experimental groups during 50 days refrigerated storage. Also, authors **Hassanzadeh *et al.* (2022)** did not report lipid protective ability of dietary astaxanthin in rainbow trout fillets. Results of TBARs assay from whole storage period are presented in Table 4.

Table 4 Results of TBARS assay (mg MDA $.kg^{-1}$)

Group	Start	Month 1	Month 2	Month 3	Month 4
Carp_0	0.293 ± 0.090^{a}	0.471 ± 0.097^{a}	0.233 ± 0.052^{a}	0.445 ± 0.223^{a}	2.176 ± 0.654^{a}
Carp_10	0.428 ± 0.150^{a}	0.388 ± 0.021^{a}	0.216 ± 0.014^{a}	0.629 ± 0.189^{a}	1.092 ± 0.146^{a}
Carp_15	0.564 ± 0.137^{a}	0.546 ± 0.165^{a}	0.261 ± 0.083^{a}	0.568 ± 0.332^{a}	1.596 ± 0.584^{a}
P-value	0.17	0.44	0.66	0.77	0.17

Note: Means in a column with a different superscript letter (a,b) are statistically different (ANOVA with Duncan test, $p<0.05$)

CONCLUSION

In our study, we incorporated astaxanthin into the diet of common carp fish to observe potential changes in oxidation stability during storage, chemical parameters, and fatty acid profile. During our experiment, we observed a significantly increased protein content in carp fish with dietary astaxanthin addition. Also, a lowering of fat content was observed. These changes, however, were not significant. Regarding oxidation stability, there were no significant differences between the control group and experimental groups during the whole storage period of 4 months. When compared fatty acid profiles of all observed common carp groups, we observe very little variance. Only significant differences were observed in stearic and arachidonic acid and only in the experimental group with lower astaxanthin addition. Based on our results, we can, therefore, conclude that dietary astaxanthin has no negative effect on fatty acid profile or oxidation stability during storage (at -18 °C). On the other hand, astaxanthin has the potential to enhance the protein and lower the fat content, even in low concentrations.

Acknowledgments: This work was supported by the Slovak Research and Development Agency under grants KEGA 001SPU-4/2023 and VEGA 1/0402/23.

REFERENCES

Abdulrahman, N. M. (2020). Effect of Algal Astaxanthin Powder Supplementation on Growth Performance, Hematological and Biochemical Parameters in Common carp, *Cyprinus carpio* L. Baerj. Com, 4(2), 116-126.

Aracati, M. F., Rodrigues, L. F., De Oliveira, S. L., Rodrigues, R. A., Conde, G., Cavalcanti, E. N. F., Borba, H., Charlie-Silva, I., Fernandes, D. C., Eto, S. F., & De Andrade Belo, M. A. (2022). Astaxanthin improves the shelf-life of tilapia fillets stored under refrigeration. *Journal of the Science of Food and Agriculture*, *102*(10), 4287–4295. https://doi.org/10.1002/jsfa.11780

Balendra, V., & Singh, S. K. (2021). Therapeutic potential of astaxanthin and superoxide dismutase in Alzheimer's disease. Open Biology, 11(6). https://doi.org/10.1098/rsob.210013

Brendler, T., & Williamson, E. M. (2019). Astaxanthin: How much is too much? A safety review. *Phytotherapy Research*, *33*(12), 3090–3111. https://doi.org/10.1002/ptr.6514

Bušová, M., Kouřimská, L., & Tuček, M. (2020). Fatty acids profile, atherogenic and thrombogenic indices in freshwater fish common carp (*Cyprinus carpio*) and rainbow trout (*Oncorhynchus mykiss*) from market chain. *Central European Journal of Public Health*, *28*(4), 313–319. https://doi.org/10.21101/cejph.a5966

Chalamaiah, M., Jyothirmayi, T., Diwan, P. V., & Kumar, B. D. (2015). Antioxidant activity and functional properties of enzymatic protein hydrolysates from common carp (*Cyprinus carpio*) roe (egg). *Journal of Food Science and Technology*, *52*(9), 5817–5825. https://doi.org/10.1007/s13197-015-1714-6

Chen, Z., Qiu, S., Amadu, A. A., Shen, Y., Wang, L., Wu, Z., & Ge, S. (2020). Simultaneous improvements on nutrient and Mg recoveries of microalgal bioremediation for municipal wastewater and nickel laterite ore wastewater. *Bioresource Technology*, *297*, 122517. https://doi.org/10.1016/j.biortech.2019.122517

Choubert, G., Mendes-Pinto, M. M., & Morais, R. (2006). Pigmenting efficacy of astaxanthin fed to rainbow trout *Oncorhynchus mykiss*: Effect of dietary astaxanthin and lipid sources. *Aquaculture*, *257*(1–4), 429–436. https://doi.org/10.1016/j.aquaculture.2006.02.055

Fajmonova, E., Zelenka, J., Komprda, T., Kladroba, D., & Sarmanova, I. (2003). Effect of sex, growth intensity and heat treatment on fatty acid composition of common carp (Cyprinus carpio) fillets. *Czech Journal of Animal Science*, *48*(2), 85–92.

Fan, K. W., & Chen, F. (2007). Production of High-Value products by Marine Microalgae thraustochytrids. In Elsevier eBooks (pp. 293–323). https://doi.org/10.1016/b978-044452114-9/50012-8

Franěk, R., Kašpar, V., Shah, M. A., Gela, D., & Pšenička, M. (2021). Production of common carp donor-derived offspring from goldfish surrogate broodstock. *Aquaculture*, *534*, 736252. https://doi.org/10.1016/j.aquaculture.2020.736252

Hassanzadeh, P., Ahmadvand, M., Aslani, S., Sheikhzadeh, N., Mousavi, S., Khatibi, S. A., Dawood, M. A., & Ahmadifar, E. (2022). Dietary astaxanthin mitigated paraquat-induced oxidative stress in rainbow trout (*Oncorhynchus mykiss*) fillet. *Aquaculture Research*, *53*(15), 5300–5309. https://doi.org/10.1111/are.16014

Jagruthi, C., Yogeshwari, G., Anbazahan, S. M., Mari, L. S. S., Arockiaraj, J., Mariappan, P., Sudhakar, G. R. L., Balasundaram, C., & Harikrishnan, R. (2014). Effect of dietary astaxanthin against Aeromonas hydrophila infection in common carp, Cyprinus carpio. Fish & Shellfish Immunology, 41(2), 674–680. https://doi.org/10.1016/j.fsi.2014.10.010

Jurčaga, L., Bobko, M., Bučko, O., Mendelová, A., Belas, P., Kročko, M., Lidiková, J., Bobková, A., Demianova, A., Poláková, K.,, & Mesarosova, A. (2022). Effect of Amelanchier extract on lipid oxidation and sensory features of pork sausages. *The Journal of Microbiology, Biotechnology and Food Sciences*, e9433. https://doi.org/10.55251/jmbfs.9433

Kojima, H., Kôda, H., Asami, S., Kiso, Y., & Tanaka, T. (2002). Contribution of the antioxidative property of astaxanthin to its protective effect on the promotion of cancer metastasis in mice treated with restraint stress. Life Sciences, 70(21), 2509–2520. https://doi.org/10.1016/s0024-3205(02)01522-9

Kumar, S., Kumar, R., Diksha, Kumari, A., & Panwar, A. (2021). Astaxanthin: A super antioxidant from microalgae and its therapeutic potential. *Journal of Basic Microbiology*, *62*(9), 1064–1082. https://doi.org/10.1002/jobm.202100391

Ljubojević, D. (2016). *Fatty acid composition, chemical composition, and processing yield of traditional hot smoked common carp (Cyprinus carpio, L)*. https://aquadocs.org/handle/1834/12116

Mráz J, Pickova J. (2011). Factors influencing fatty acid composition of common carp (*Cyprinus carpio*) muscle. *Neuro Endocrinol Lett. Suppl* 2:3-8. Mráz, J., & Pickova, J. (2011). Factors influencing fatty acid composition of common carp (Cyprinus carpio) muscle. PubMed, 32 Suppl 2, 3–8. https://pubmed.ncbi.nlm.nih.gov/22101873

Park, J. S., Mathison, B., Hayek, M. G., Massimino, S., Reinhart, G. A., & Chew, B. P. (2011). Astaxanthin stimulates cell-mediated and humoral immune responses in cats. Veterinary Immunology and Immunopathology, 144(3–4), 455–461. https://doi.org/10.1016/j.vetimm.2011.08.019

Poláková, K., Bobková, A., Demianová, A., Bobko, M., Lidiková, J., Jurčaga, L., Belej, Ľ., Mesárošová, A., Korčok, M., & Tóth, T. (2023). Quality attributes and sensory acceptance of different botanical coffee Co-Products. *Foods*, *12*(14), 2675. https://doi.org/10.3390/foods12142675

Shah, M. M. R., Liang, Y., Ji, C., & Daroch, M. (2016). Astaxanthin-Producing Green Microalga Haematococcus pluvialis: From Single Cell to High Value Commercial Products. *Frontiers in Plant Science*, *7*. https://doi.org/10.3389/fpls.2016.00531

Song, X., Wang, L., Li, X., Chen, Z., Liang, G., & Leng, X. (2016). Dietary astaxanthin improved the body pigmentation and antioxidant function, but not the growth of discus fish (Symphysodon spp.). Aquaculture Research, 48(4), 1359–1367. https://doi.org/10.1111/are.13200

Stachowiak, B., & Szulc, P. (2021). Astaxanthin for the food industry. Molecules, 26(9), 2666. https://doi.org/10.3390/molecules26092666

Trembecká, L., Haščík, P., Čuboň, J., Bobko, M., & Pavelková, A. (2016). Fatty acids profile of breast and thigh muscles of broiler chickens fed diets with propolis and probiotics. *Journal of Central European Agriculture*, *17*(4), 1179–1193. https://doi.org/10.5513/jcea01/17.4.1828

Yeganeh, S., Shabanpour, B., Hosseini, H., Imanpour, M. R., & Shabani, A. (2012). Comparison of farmed and wild common carp (*Cyprinus carpio*): Seasonal variations in chemical composition and fatty acid profile. *Czech Journal of Food Sciences*, *30*(6), 503–511. https://doi.org/10.17221/455/2011-cjfs

Zhu, X., Hao, R., Zhang, J., Tian, C., Yuan, H., Zhu, C., & Li, G. (2022). Dietary astaxanthin improves the antioxidant capacity, immunity and disease resistance of coral trout (Plectropomus leopardus). Fish & Shellfish Immunology, 122, 38–47. https://doi.org/10.1016/j.fsi.2022.01.037