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About the Journal

Focus and Scope

Pharmacy Education journal provides a research, development and evaluation forum for communication between academic teachers, researchers and practitioners in professional and pharmacy education, with an emphasis on new and established teaching and learning methods, new curriculum and syllabus directions, educational outcomes, guidance on structuring courses and assessing achievement, and workforce development. It is a peer-reviewed online open access platform for the dissemination of new ideas in professional pharmacy education and workforce development. Pharmacy Education supports Open Access (OA): free, unrestricted online access to research outputs. Readers are able to access the Journal and individual published articles for free - there are no subscription fees or 'pay per view' charges. Authors wishing to publish their work in Pharmacy Education do so without incurring any financial costs.

In addition we are listed in EBSCO, and indexed in the Emerging Sources Citation Index (ESCI - Web of Science), EMBASE and SCOPUS.

The Journal also recognises the importance of policy issues and current trends in the context of education, professional development and workforce.

The Journal publishes reports of research and innovation in all aspects of professional pharmacy education and training, case studies, country studies, innovations in laboratory and professional educational practice, workforce issues and development, reviews and reports on information technology in education and reviews of current literature.

The Journal has a clear international perspective, and has a longstanding policy of facilitating publication, in particular for younger Faculty, and those authors whose first language may not be English, and manuscripts from all regions seeking low cost engagement with the wider global community.

The Journal is published by the International Pharmaceutical Federation (FIP) and is aligned to the global mission of advancing education, advancing practice and advancing science.

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Pharmacy Education has adopted a double-blind peer review process - the identities of the Authors and Reviewers are kept from being known to each other. A step-by-step checklist is provided for Authors, Reviewers and Editors to ensure this (see Ensuring a Blind Review).

Peer Review Process: Once a submission is received, the assigned Editor will select appropriate Reviewers based on their expertise and proven ability to critique. The peer reviews received will assist the Editor in determining the validity, significance and originality of the work submitted. Reviewers will also provide comment on manuscript content for scientific value, check for adherence to general scientific practice as well as Pharmacy Education's specific guidelines. The Peer Review process will look closely at methodology and the data validity, and consider the ethical approach. Reviewers are encouraged to provide suggestions for improvement and recommend to Editors if manuscripts should be accepted, accepted with revisions, or rejected.

Please note that an invitation for Authors to submit a revised version is not a guarantee of acceptance. Ultimately, the final decision lies with the Editor assigned to each submission. An Editor can reject any article at any time before publication, including after acceptance if concerns arise about the integrity of the work.

As part of their agreement with Pharmacy Education, Reviewers will keep manuscripts and associated material strictly confidential, and will not appropriate Authors' ideas before the manuscript is published. Once a review has been completed, Reviewers will be directed and expected to permanently delete/destroy any retained copies of manuscripts they hold (see Privacy Statement).

Timeliness: Reviewers are expected to respond promptly to requests to review and to submit reviews within the time agreed. Reviewers are also required to declare their conflicts of interest and recuse themselves from the Peer Review process if a conflict exists. Editors will do their utmost to ensure timely processing of manuscripts. Authors will be notified on any unusual delays in publication of manuscripts via email. Authors will ne notified as soon as possible if a manuscript is going to be rejected, either by the Journal Manager or Editorial Team.

Journal Ownership and Editorial Scope

Pharmacy Education is published by the International Pharmaceutical Federation (FIP). Appointments and dismissals to the Editorial Team are made by the Editor-in-Chief in consultation with FIP.

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Editor-in-chief - The Editor-in-Chief has full authority over content publication in Pharmacy Education. In co-operation with the wider Editorial Team and publisher, they direct overall strategy of the journal. Together with the Editors and Associate Editors, the Editor-in-Chief reviews and decides upon submitted manuscripts, ensuring timely publication of submissions.

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Advisory Board – Pharmacy Education is currently engaged in establishing an Advisory Board who alongside the Editor-in-Chief, Editors and Associate Editors will assist with:

- Guidance on the peer review and publishing policies of Pharmacy Education and where necessary, suggest reviewers to the Editor-in-Chief.
- Developing the journal by providing expertise to the Editor-in-Chief and FIP on how to increase impact and reach
- Impartial Judgement in appeal cases by providing professional, independent scientific comments to the Editor-in-Chief and FIP
- Promoting Pharmacy Education

Managing Editor – The Managing Editor assumes day-to-day responsibility of managing the submissions flow to Pharmacy Education. They liaise with Authors and Reviewers where needed, clarifying the Submission and Publication process as well as responding to all general enquires. The Managing Editor also completes all typesetting, proofreading and online publication of accepted manuscripts once accepted by the Editors.

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Peer Reviewers: Reviewers are asked at the time of conducting a review if they have conflicts of interest that may impact on their ability to provide an unbiased review. Reviewers are asked to disclose conflicts of interest to the assigned Editor. The assigned Editor will then cancel the review and reassign the article to another reviewer. Reviewers agree to not use knowledge of the work they are reviewing before its publication to further their own interests.

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Research involving Human participants and Informed Consent

It is the responsibility of the authors to ensure that research involving human subjects has been reviewed and approved by the appropriate research or ethics review committee, or that it has been determined to be exempt from such review.

Confirmation of this should be included in the Cover Letter and also included in the Methods section of the manuscript. Where informed consent is required, authors should include a statement in the manuscript detailing that informed consent was obtained from human subjects (see Submission Preparation Checklist)

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Published articles are a permanent record that should remain unaltered. However, Pharmacy Education recognises that in exceptional circumstances, articles may need to be corrected, replaced, retracted or removed.

The Editor-in-Chief has full authority over content publication in Pharmacy Education. In making decisions regarding publication, the Editor-in-Chief is guided by the policies of the Journal as well as legal requirements such as libel, copyright, infringement and plagiarism.

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Detailed below are our procedures for managing requests for corrections post publication

Minor errors

If Authors identify a minor error once an article has been published online, they are advised to email their request for corrections to Pharmacy Education for consideration.

Minor errors include: errors in spelling, data, medical terms; missing text; amendments to tables, figures or appendices; errors in correspondence details, etc. The Journal may decline proposed corrections that are for aesthetic reasons; errors to text, typography tables, figures and appendices if the meaning is unchanged; errors in acknowledgments lists etc.

Significant Corrections

Corrections may be needed if honest errors have resulted in a portion of an article being misleading; if the author/contributor lists are disputed; or if potential conflicts of interest affecting authorship are disclosed post publication.

Where the Editor-in-Chief agrees that a correction is needed, the Journal will:

- Correct the error online, and to any article file for download, linking to a Correction Notice via a footnote
- The Correction Notice will detail the changes made to the original version, and the dates the changes were made.

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Honest errors such as mis-classification or miscalculation may lead to significant changes to the results, interpretations and conclusions. In such cases, the Journal will consider retraction with replacement of the article:

- The changed version of the article will undergo further editorial review;
- The authors will be required to detail and explain the changes made which will be published as supplementary material or in an appendix;
- The supplementary material/appendix will be attached to the changed version, allowing for complete transparency.

Retraction

An article will be retracted if the results or conclusions are unsound and/or where misconduct breaching professional ethical codes has occurred. The publisher and Editor-in-Chief will conduct an investigation into the errors or misconduct before retracting an article. The following steps will be taken where articles are retracted:

- A Statement of Retraction, giving the reasons for the retraction and signed by the authors and/or the Editor-in-Chief will be published online linking to the original article.
- The original article is preceded by a screen containing the Statement of Retraction. The reader can then proceed to the article itself.
- A watermark will be added to the original PDF indicating on each page that it is “RETRACTED”
- The Statement of Retraction will be included as a numbered page in the Table of Contents to ensure proper indexing, and will include the article title in its heading

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Very occasionally, it may be necessary to remove an article from the online database as a consequence of legal action (e.g., defamatory content, infringement on legal rights, article is subject of a court order, or might pose a serious health risk if an article's content is acted upon).

In these circumstances:

- The article's metadata (title and author details) will be retained and the text replaced with an Article Removal Notice
- The Article Removal Notice will be included in the Table of Contents and prefix the metadata.

Expressions of Concern

If concerns or allegations of misconduct regarding a publication are raised, the Editor-In-Chief will consult the Committee on Publication Ethics (COPE) http://www.publicationethics.org and initiate the appropriate procedure based on the nature of the concern or allegation. The Editor-in-Chief, with appropriate support form the Editorial Team, will assess each situation individually.

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- investigations into any concerns of misconduct remain inconclusive;
- concerns remain over the impartiality of any investigations into alleged misconduct;
- -an investigation is pending and a judgment is not expected for some time.

An Expression of Concern will be published and appear in the Table of Contents and include the title of the article in its heading. It should be noted that Pharmacy Education understands the potential repercussions that issuing an Expression of Concern can bring and will only take this action where it is deemed necessary.

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Authors are entitled to appeal editorial decisions if they believe their submission has been unfairly or inappropriately rejected. An appeal letter should be submitted to the Journal Manager (pej@fip.com)

The appeal letter should provide appropriate detail and context. For example, if an Editor has provided peer review comments it is worthwhile responding to each item in the letter. If the appeal is against the editorial decision made on the submission, explaining and justifying clearly the work's importance, relevance, and usefulness in the appeal letter is recommended.

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- the complainant defines their dissatisfaction as a complaint; and
- it concerns a failure of process, i.e. a long delay or a severe misjudgement; and is not simply disagreement with an editorial decision;
- the issue being raised is within the responsibility of Pharmacy Education's editorial remit
- Complaints should be directly emailed to the Journal Manager (pej@fip.com) who will ordinarily formally acknowledge receipt within 5 working days.
- The Journal Manager will forward the complaint to a relevant person within the Journal organisation who will aim to provide a full response within four weeks. If this is not possible, an interim response and update will be given within the four weeks.
- Following this action, if the complainant remains unhappy, complaints will be escalated to the Editor-in-Chief whose decision is final.
- If a complainant remains unhappy, they may complain to an external body such as the Committee on Publication Ethics (COPE) http://www.publicationethics.org. They will consider complaints against Editors once a Journal's own Complaints procedures have been exhausted.

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Journal History

Pharmacy Education has been publishing peer reviewed education, training, research and evaluation in the field of pharmaceutical education since 2000.

The Journal encourages manuscript submissions from younger career scientists, academics and practitioners and has a focus on supporting authors who do not have English as a first language.

Through our FIP publication platform we are able to reach out to over 3 million pharmacists and pharmaceutical scientist worldwide.

Editorial Team

Vol. 21 No. 2 (2021): IAI Conference 2020

We are pleased to confirm the publication of IAI Conference 2020.

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Conference Proceedings

IAI CONFERENCE: The therapeutic outcomes and adverse drug reactions study of Clozapine on Schizophrenia inpatients in the Grhasia psychiatric hospital Yogyakarta, Indonesia

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IAI CONFERENCE

RESEARCH ARTICLE

The potential effect of the green coffee extract on reducing atherogenic index in hyperlipidemic rats

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Abstract

Introductions: Dyslipidemia is a risk factor for atherosclerosis and cardiovascular disease. The high prevalence of dyslipidemia triggers the development of green coffee supplement products, which are claimed as cholesterol-lowering and slimming agents. Nonetheless, research data on the effect of taking green coffee supplement products, especially regarding cardiovascular function, is limited. **Aims:** To determine the potential effect of green coffee extract (GCE) on improving atherogenic index of plasma (AIP) and cardiac histopathology in hyperlipidemic rats. **Methods:** 24 rats were induced by high-fat feed for 21 days. Then, the rats were treated with a GCE, dose of 200, 400, and 800 mg/kg bodyweight for 14 days. The next day, blood was collected from the rats to take measurements of their serum lipid profile and calculating their AIP. The heart organ was created by using histopathological preparations. **Results:** Administration of GCE in all doses significantly reduced the AIP and improved cardiac histopathology in the hyperlipidemic rats. **Conclusions:** GCE can be developed as a cardio-protector.

Introduction

Dyslipidemia is a significant fat component disorder, which includes an increase in total cholesterol and Low-Density Lipoprotein (LDL) levels (called hypercholesterolemia), an increase in triglyceride levels (called hypertriglyceridemia), and a decrease in High-Density Lipoprotein (HDL) levels, or a combination thereof (DiPiro *et al*., 2014). This abnormal condition needs special attention because it is a risk factor for atherosclerosis which will lead to cardiovascular disease that comes from narrowing of blood vessels due to fat accumulation (Nelson, 2013). This disease caused by excess fat is a fairly common health problem.

In 2008, about 39% of 25-year-old had increased cholesterol levels globally. This increase in cholesterol was estimated to be the cause of 18% of cerebrovascular disease cases and 56% of ischemic heart disease in the world. The death rate caused by dyslipidemia disease affects 4.4 million people (WHO, 2002). Based on the 2013 Riskesdas Report, for people aged below 15 years old, there was 35.9% abnormal total cholesterol, 22.9% low HDL, 60.3% optimal-borderline high LDL and 15.9% high-very high LDL, abnormal triglycerides with high borderline categories of 13.0% and high-very high categories of 11.9%. East Java is in the top six for obesity cases and second place after DI Yogyakarta with a cardiovascular disease prevalence of 0.2% (Ministry of Health, 2013).

Certain factors can trigger the high prevalence of dyslipidemia to cause serious diseases. The interaction of genetic factors and environmental factors can cause hyperlipidemia (PERKI, 2013). Unhealthy lifestyles prevalent in today's society can also trigger fat accumulation in the blood circulation; these include the penchant for consuming food from fast-food restaurants and other fat-rich foods, lack of activity and exercise, alcohol consumption, obesity and a family history of hyperlipidemia (LIPI, 2009). Generally, this

disease does not cause symptoms in sufferers, but it was found that some patients had symptoms of chest pains, anxiety, sweating, and shortness of breath (DiPiro *et al*., 2014).

Dyslipidemia is one of the risk factors associated with coronary heart disease. Low HDL levels, as well as high levels of triglycerides and LDL, correlate with an increased incidence of coronary heart disease. Several lipid ratio parameters, such as the atherogenic index of plasma (AIP), Castelli's Risk Index, and the atherogenic coefficient, can be used to predict the risk of cardiovascular disease (Bhardwaj *et al*., 2013). Controlling these various lipid ratios, hopefully, can help the prevention of cardiovascular disease events and the management of dyslipidemia therapy.

In addition to the primary therapy with the statin class and lifestyle modification (LIPI, 2009; Walker, & Whittlesea, 2012), recently there are several products in the form of cholesterol-lowering supplements, slimming or weight loss on the market to reduce fat in the body. One of them is green coffee extract supplements. Green coffee is coffee that has not undergone a roasting process like black coffee and is known to have a higher chlorogenic acid content (Farah, 2012). It is also supported by the fact that Jember city is one of the five most significant contributors to Robusta coffee in East Java (Ministry of Agriculture, 2016).

Several studies have shown that the active compounds in coffee have beneficial effects, such as antioxidant (Sato *et al*., 2011), hepatoprotective (Ji *et al*., 2013), and antidiabetic effects (Ong *et al*., 2013). Besides, the active compounds in coffee can prevent the storage of carbohydrates and lipids (Shimoda *et al*., 2006). Chlorogenic acid in green coffee can increase the oxidation of fatty acids (Li *et al*., 2009). Research conducted by Shimoda and colleagues (2006) and Choi and colleagues (2016) proved that green coffee bean extract could cause weight loss in mice. Regarding lipid profile parameters, giving green coffee ethanol extract can significantly reduce LDL levels, increase HDL, and significantly reduce total cholesterol levels in white rats when induced with a high-fat diet (Setyono *et al*., 2014). Other research stated that green coffee extract could lower cholesterol and lower total triglyceride levels (Choi *et al*., 2016). Meanwhile, the latest study showed that administration of green coffee extract for 14 days in hyperlipidemic rats could significantly improve lipid profiles, except HDL levels which remained unchanged. It also enhanced the rat aorta histopathology even though it did not match normal conditions (Christianty *et al*., 2020).

So far, however, there has been no research data on the activity of green coffee extracts related to lipid ratio parameters to predict cardiovascular events. Therefore, this study aimed to determine the potential of green coffee extract in preventing cardiovascular disease based on atherogenic index parameters and histopathological cardiac features of the hyperlipidemic rats.

Materials and methods

Materials

Green bean coffee as a sample was obtained from KSU Buah Ketakasi, Sidomulyo, Silo District, Jember City. The materials used in the study included ethanol, aqua dest, HCl_2N, normal saline, NH_4OH, simvastatin tablet 10 mg (PT. Hexpharm Jaya), CMC Na, used cooking oil, quail egg yolk, BR II feed (PT Japfa Comfeed), formaldehyde, ether, triglyceride and HDL reagents, hematoxylin-eosin staining, enthelan, xylol. The experimental animals used were male Wistar rats, healthy, weight 150-250 g in weight, and around six to eight weeks old.

Methods

Preparation of green coffee extract

The green coffee extract was prepared by the maceration method. Dried green coffee beans were ground to a Simplicia powder, weighed in a certain amount and macerated using 96% ethanol 7.5 times the weight of powder. The resulting filtrate was concentrated using a rotary evaporator and then dried using an oven at a temperature of 50°C until a constant weight was obtained. Furthermore, the green coffee extract suspension was made in CMC Na 1% at a dose of 200 mg/kg, 400 mg/kg, and 800 mg/kg body weight.

In vivo activity assay

The whole procedure for the care and treatment of experimental animals had obtained the approval of the ethics committee of the Jember State Polytechnic with certificate number 615/PL17/LL/2018. A total of 25 rats were acclimatised first for seven days, then administered a high-fat diet, with a composition of quail egg yolk, using cooking oil and 0.01% propylthiouracil (PTU) orally for 21 days, except in the control group. Hyperlipidemic rats were then divided into several groups, namely the green coffee extract treatment group (dose 200 mg/kg, 400 mg/kg and 800 mg/kg body weight), and the positive control group was given simvastatin suspension 0.9 mg/kg BW and CMC Na 1% for negative control. All treatments were given orally for 14 days. On the 15th day, the rats were killed, intracardiac blood was taken for measurement of lipid

profiles, and the heart organs were separated to make HE preparations.

Lipid profile measurements

The lipid profile measured was triglyceride and HDL levels. Triglyceride levels were measured using the enzymatic colourimetric method using glycerol-3-phosphate-oxidase (GPO-PAP). The test was started by mixing 10 µL of serum with 1000 µL of reagent and then incubated at 37°C for five minutes. The 1000 µL reagent mixture and 10 µL standard solution were treated the same, and then the absorbance was measured using a 546 nm wavelength Biolyzer-100TM photometer.

HDL levels were measured by the precipitation method and determined enzymatically. A sample of 200 µL was mixed with 500µL of HDL reagent, left to stand for 10 minutes at 15-25^{o} C, then centrifuged ten minutes at 4000g. The supernatant was removed from the residue within one hour. Next, pipette 100µL supernatant and mix with 1000 µL cholesterol reagent, incubated at 37^{o}C for five minutes. For the control, use 100 µL of aqua bidest with 1000 µL of cholesterol reagent, treated the same as the sample. The absorbance of the sample was measured within 60 minutes using a 546 nm wavelength Biolyzer-100TM photometer.

Atherogenic index determination

The atherogenic index was calculated based on the logarithm of the ratio between triglyceride levels and HDL levels in molar units (mmol/L), with the following formula (Frohlich & Dobiasova, 2003): Atherogenic index of plasma (AIP) = Log (TG/HDL)

Statistical analysis

The lipid profile data and AIP were expressed as means ± standard deviation ($\bar{x}$ ± SD). Between-group comparisons were performed using one-way analysis of variance (ANOVA), followed by the Least Significant Difference (LSD) procedure for multiple range tests. A value of $p < 0.05$ was considered significant.

Results

In this study, the induction of high-fat feed used a combination of quail egg yolk and used cooking oil (7:3) which was administered once a day, and 0.01% PTU suspension in distilled water given ad libitum. The induction was carried out for three weeks, and the results of the increase in lipid profile were obtained.

Based on the results shown in Table I, it is found that the triglyceride levels in the treatment group (both with green coffee extract and simvastatin) are lower than the negative control group. The results of statistical analysis using one-way ANOVA showed that there were significant differences ($p = 0.023$) between groups. The post hoc test with LSD showed that administration with green coffee extract had triglyceride levels that were significantly different from the CMC Na 1% group ($p < 0.05$) but not separate from the simvastatin group ($p > 0.05$). Between groups, the dose of the green coffee extract also did not show a significant difference. It means that treatment with green coffee extract (starting at a dose of 200mg/kg BW) can reduce triglyceride levels in hyperlipidemic rats, and it is equivalent to 0.9mg/kg BW of simvastatin. However, this was not the case for HDL levels, which between groups did not show a significant difference ($p = 0.379$).

The lipid profile (especially triglyceride and HDL level) data was then used to determine the AIP. Table I shows that the treatment with green coffee extracts for 14 days can reduce the AIP. According to one-way ANOVA analysis, as written in Table I, the AIP between groups has a significant difference ($p = 0.007$). The post hoc test with LSD showed that the green coffee extract group at various doses was significantly different from the CMC Na 1% group but not significantly different when compared to the simvastatin control group. Among the three doses of green coffee extract, the atherogenic index value was not significantly different. It means that green coffee extract (starting at a dose of 200 mg/kg BW) can reduce the AIP in hyperlipidemic rats, equivalent ability with 0.9 mg/kg BW simvastatin. However, based on the atherogenic index risk category classification, namely low (< 0.11), moderate (0.11 - 0.21), and high (> 0.21) (Dobiasova *et al.*, 2004), it was found that all groups had a low-risk category.

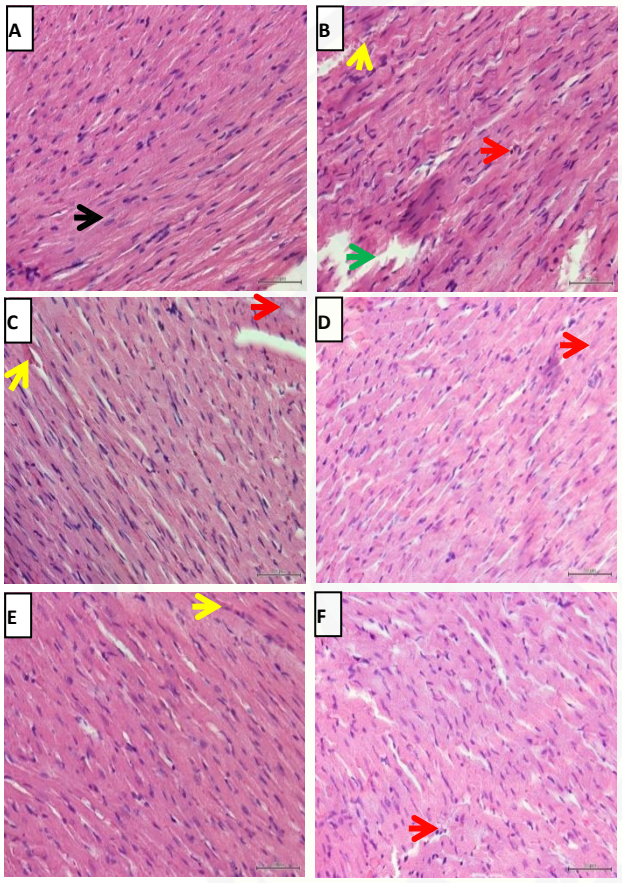
Table I: Profile of triglyceride and HDL level, and atherogenic index of plasma (AIP)

Groups	Mean level (mmol/L) ± SD		AIP	Interpretation (Based on risk category)
	Triglyceride	HDL		
Normal	0.61±0.32^{a}	0.91±0.20	-0.21±0.17^{a}	Low
CMC Na 1%	1.18±0.33^{b}	1.05±0.32	0.10±0.14^{b}	Low
Simvastatin	0.67±0.21^{a}	1.43±0.48	-0.33±0.19^{a}	Low
GCE 200 mg/kg bw	0.79±0.12^{a}	1.04±0.39	-0.15±0.19^{a}	Low
GCE 400 mg/kg bw	0.51±0.13^{a}	1.34±0.22	-0.42±0.06^{a}	Low
GCE 800 mg/kg bw	0.78±0.29^{a}	1.06±0.26	-0.18±0.21^{a}	Low

Different superscript letters indicate that there are significant differences between groups ($p < 0.05$); Anova test followed by LSD; GCE = Green coffee extract

The administration with green coffee extract and simvastatin could improve the cardiac histopathological feature of hyperlipidemic rats on

microscope observation. In normal rats, myocyte cells (longitudinal/elongated muscle fibres) were neatly arranged, parallel, and normal cell nucleus (→) (Fig. 1A). In the high-fat diet-induced rat, there were structural changes/degeneration in myocytes (→), vascular dilatation and congestion (→), and necrosis (→) (Figure 1B, 1C, 1D, 1E, 1F). The necrosis shown included pyknosis (cell nucleus shrinkage), karyorrhexis (destroyed cell nuclei), and karyolysis (cell nuclei disappeared). There was an improvement in the histopathological picture of cardiac myocytes in all treatment groups. Although some cells experienced necrosis and congestion, there was no degeneration of myocytes in the treatment group.



A. Normal rat heart muscle cells (myocytes), B. negative control (CMC Na 1%), C. positive control (simvastatin 0.9 mg/kg BW), D. GCE 200 mg/kg BW, E. GCE 400 mg/kg BW, F. GCE 800 mg/kg BW, → normal myocytes, → myocyte degeneration, → cell necrosis, → vascular dilatation and congestion.

Figure 1: Histopathology of rat heart, a cross-section with hematoxylin-eosin staining, magnification 400x

Discussion

The induction of high-fat feed used a combination of quail egg yolk and used cooking oil (7:3) which was administered once a day, and 0.01% PTU suspension in distilled water given ad libitum for three weeks successfully made hyperlipidemic model in rats. Quail egg yolk has the highest cholesterol content compared to other egg yolks, namely 2139.17 mg/100 g of eggs (Dwiloka, 2003; Aziz *et al.*, 2012). Used cooking oil contains saturated fatty acids and can increase the number of cells experiencing necrosis in rat hearts (Nurfadilah *et al.*, 2013). The addition of PTU is intended to damage the thyroid gland. Hypothyroidism can cause a decrease in the synthesis and expression of LDL receptors in the liver. That makes LDL circulate a lot in the plasma and drives hypercholesterolemia (Kapourchali *et al.*, 2014).

The green coffee extract could reduce the triglyceride level, but not with an HDL level. These results are in line with previous research, where green coffee extract could decrease all lipid profiles in high-fat feed induced rats, except HDL (Christianty *et al.*, 2020). Another study showed that the ethanol extract of green coffee beans at doses of 200 and 400 mg/kg BW could significantly reduce triglyceride levels (Shimoda *et al.*, 2006). A slightly different result was found in the study conducted by Setyono and colleagues (2014), where giving green coffee ethanol extract at a dose of 400 mg/kg BW was more efficient in increasing HDL significantly in white rats induced by a high-fat diet. Still, there was no difference in lowering triglycerides (Setyono *et al.*, 2014). This difference is influenced by its habitat and the processing of coffee. The difference in altitude where it grows will also affect the levels of active compounds in a plant, as well as the processing.

The increase in triglyceride levels is influenced by the rise in energy intake from the high-fat feed given. It can increase the activity of lipogenesis so that more free fatty acids are formed. The mobilisation of free fatty acids to the liver will also increase, and then it will be esterified with glycerol to form triglycerides. Meanwhile, HDL levels are more influenced by triglyceride metabolism due to lipoprotein lipase activity (Rodwell *et al.*, 2018).

The green coffee extract could also lower the index atherogenic. Based on lipid profile, only the triglyceride level was affected due to differences in treatment, whereas HDL was not. Of course, it will affect the AIP, which is the result of the ratio logarithm of triglyceride and HDL levels. The higher the triglyceride level, the greater the atherogenic index value. Otherwise, the higher the HDL level, the smaller the AIP value. The AIP describes the distribution of lipids and lipoproteins in the body, which correlates significantly with the presence of risk factors for atherosclerosis, such as gender, age, dyslipidemia, and diabetes, as well as coronary angiography (Frohlich & Dobiasova, 2003). Also, the atherogenic index can be used as a parameter for routine daily monitoring, especially in patients with

other risk factors for cardiovascular disease (Niroumand *et al*., 2015). Compared to different parameters, the AIP has the most excellent sensitivity (84%) for predicting atherogenicity and the incidence of cardiovascular disease (Khazaál, 2013) and has the largest correlation coefficient compared to other lipid ratio parameters to the incidence of coronary artery disease (Bhardwaj, 2013).

From the results above, all of the groups are at low risk of developing a cardiovascular event. This was potentially due to the introduction of high-fat feed for only 21 days. The low atherogenic index in this study suggests that the risk of developing atherosclerosis and cardiovascular disease is still lacking.

The previous study showed that green coffee was known to improve inflammation and abnormalities in the heart, liver, and diastolic stiffness without increasing glucose sensitivity or lipid profiles in rats with metabolic syndrome (Bhandarkar *et al*., 2019a). Chlorogenic acid, as the main component of green coffee, can improve left ventricular diastolic stiffness by reducing collagen deposition and inflammatory cell infiltration in the left ventricle in high-fat feed rats (Bhandarkar *et al*., 2019b) and reducing interstitial collagen accumulation of the heart in an isoproterenol-induced myocardial infarction model in rats (Akila *et al*., 2017).

The AIP and cardiac histopathological feature improvement were presumed due to the main active compound in green coffee, namely chlorogenic acid. This compound is known to have a primary hypocholesterolemic effect and secondary effects such as atheroscleroprotective, cardioprotective, and hepatoprotective. The mechanism was presumed to increase the use of fatty acids in the liver through upregulation of peroxisome proliferation-activated receptor α (PPAR α) mRNA (Wan *et al*., 2012). Also, chlorogenic acid can activate AMPK (Ong *et al*., 2013), which causes metabolic responses, such as inhibiting fatty acid synthesis (through inhibition of fatty acid synthase) and increasing fatty acid oxidation (by increasing carnitine palmitoyltransferase (CPT) activity), inhibits lipolysis (through inhibition hormone-sensitive lipase (HSL)), and triglyceride synthesis (Meng *et al*., 2013).

Conclusion

This study concluded that green coffee extract has the potential to be developed as a protective agent against cardiovascular function through a series of advanced preclinical and clinical studies.

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