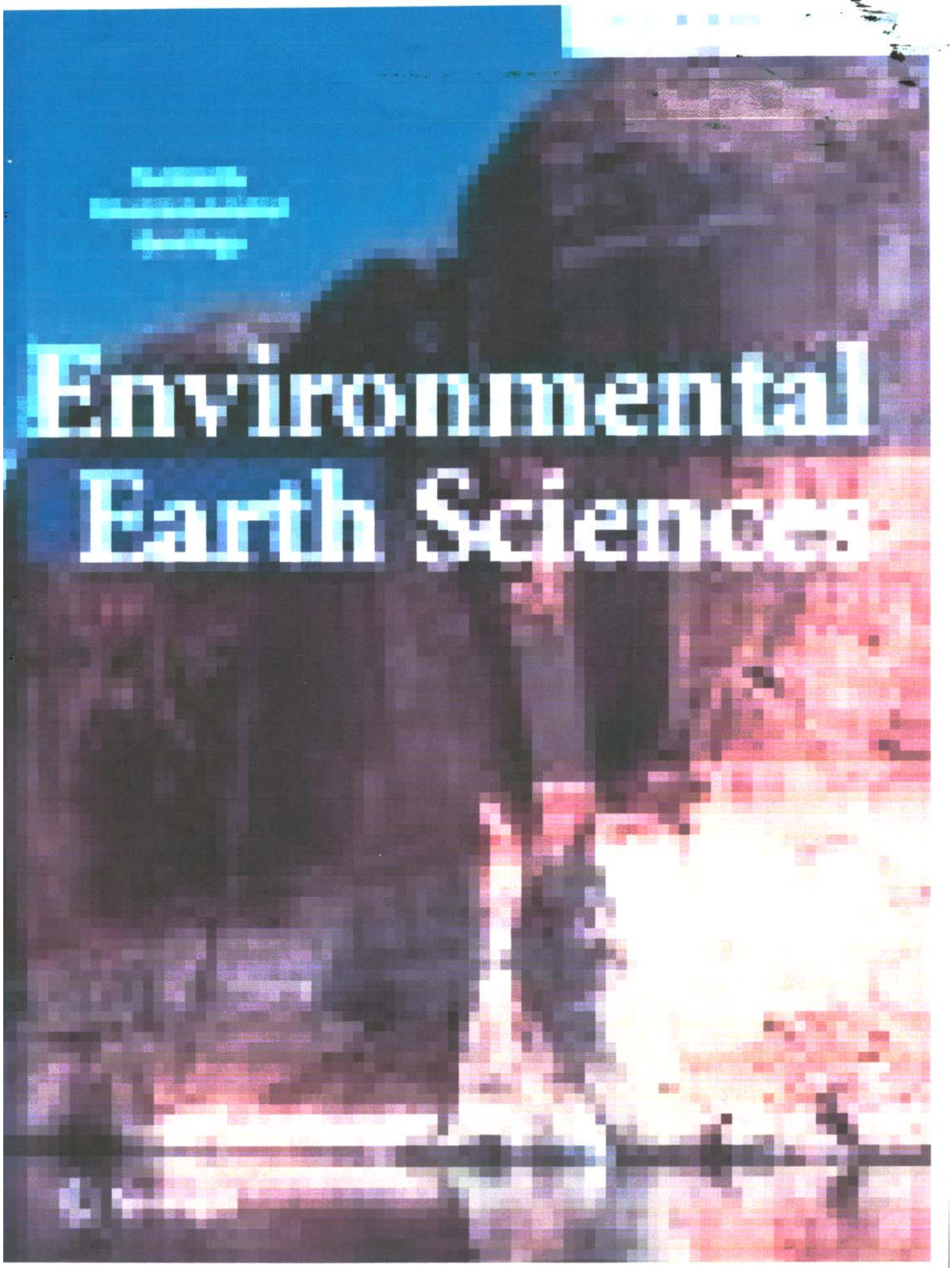

Environmental
Earth Science

Editors

National Research Council of Italy
Inst. for Agricultural and Forest Systems
in the Mediterranean (ISAFOM)
Via Cavour, 4-6
87036 Rende (CS), Italy
E-Mail: gabriele.buttafuoco@cnr.it

PETER DIETRICH
Editor - Environmental Geophysics/Environmental Monitoring
Helmholtz Centre for Environmental Research- UFZ
Dept. Monitoring and Exploration Technologies
Permoserstr. 15
04318 Leipzig, Germany
Tel: +01 0341 235 1281
E-mail: peter.dietrich@ufz.de

DEREK KIM
Editor - Water and Waste Management and Remediation
2575 Coventry Circle
Fullerton, CA 92833
USA
E-mail: Derekkim10@yahoo.com

BRODER J MERKEL
Editor - Hydrogeology and Mining Geology
TU Bergakademie Freiberg
Dept. for Geology
Gustav-Zeuner-Str. 12
09596 Freiberg/Sa.
E-mail: merkel@geo.tu-freiberg.de

JAN SCHWARZBAUER
Editor - Geochemistry
Inst. for Geology and Geochemistry
of Petroleum and Coal
RWTH Aachen University

Lochnerstr. 4-20
52056 Aachen
Germany
Tel: +49 241 8095750
Fax: +49 241 8092152
E-mail: jan.schwarzbauer@emr.rwth-aachen.de

WANFANG ZHOU
Knoxville, Tennessee 37934, USA

The effect of combination *Bifidobacterium sp* and *Lactobacillus acidophilus* probiotic on egg yolk cholesterol, HDL, and LDL

IOP Conf. Series: Earth and Environmental Science **441** (2020) 012049 doi:10.1088/1755-1315/441/1/012049

The effect of combination *Bifidobacterium sp* and *Lactobacillus acidophilus* probiotic on egg yolk cholesterol, HDL, and LDL

W P Lokapirnasari[1,6], A M Sahidu[2], L Maslachah[3], A B Yulianto[4] and R Najwan[5]

[1]Department of Animal Husbandry, Faculty of Veterinary Medicine, Jl. Mulyorejo, Kampus C, Universitas Airlangga, Surabaya, Indonesia
[2]Department of Marine, Faculty of Fisheries and Marine, Universitas Airlangga, Surabaya, Indonesia
[3] Department of Basic Medicine, Veterinary Pharmacy Laboratory, Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya, Indonesia
[4]Sains Veteriner, Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya, Indonesia
[5]Agribusiness Veteriner Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya, Indonesia

[6] Corresponding author: widyaparamitalokapirnasari@gmail.com

Abstract. The purpose of this study to evaluate the effects of combination 0.5% *Bifidobacterium* sp.+0.5% *Lactobacillus acidophilus* on egg yolk cholesterol, High density lipoprotein (HDL) and low density lipoprotein (LDL) in hens. Methods: this research using completely randomized design. Sixty layers were randomized into three treatments, each treatment contains four replicates and each replicates contains five hens. Twenty five week-old Isa Brown layers were fed without probiotic (T0), 0.1% antibiotic growth promoters (T1), 0.5% *Bifidobacterium* sp.+0.5% *L. acidophilus* (T2) for 30 days. Result: the result showed that there was a significant difference between the control and all treatment groups on egg yolk cholesterol, HDL and LDL. Supplementation of 0.5% *Bifidobacterium* sp.+0.5% *L. acidophilus* probiotic decreased the egg yolk cholesterol and LDL ($p<0.05$) and increased HDL in egg yolk ($p<0.05$) levels. The lowest egg yolk cholesterol and LDL by supplementation 0.5% *Bifidobacterium* sp.+0.5% *L. acidophilus* probiotic (155.86 and 48.46 mg/dL) respectively, compared the control (224.0200 and 58.6300 mg/dL) and the highest egg yolk HDL by supplementation of 0.5% *Bifidobacterium* sp.+0.5% *L. acidophilus* probiotic (50.00 mg/dL) compared the control treatment (41.5900 mg/dL). The conclusion of this study was the supplementation of 0.5% *Bifidobacterium* sp.+0.5% *L. acidophilus* could improve egg yolk quality of hens.

1. Introduction

Probiotics or direct-fed microbials are live microorganisms which beneficially affect the host animal by improving their intestinal microflora [1] and they were living symbiotically in the digestive tract of poultry which is known to increase host immunity through control of pathogenic bacteria [2] by improving their intestinal microflora [3]. The supplementation of probiotics has been found could increase the egg production, egg weight, and feed efficiency in laying hens [4, 5, 6]. Ishihara reported

that an increase in the amount of Lactobacillus spp in laying hens is beneficial in preventing pathogenic diseases [7].

The characterize of probiotic has adhesive properties and can colonize different parts of the avian digestive tract [8]. Lactic acid bacteria, such as *Pediococcus acidilactici, Lactobacillus* spp., *Lactococcus lactis* can produce lactic acid from the results of glucose metabolism, which serves to acidify the digestive environment, and bactericide effect that can inhibit pathogenic bacteria growth such as *E. coli*, Salmonella Typhimurium [9]. Manipulation of intestinal microbial with probiotics plays an important role in the lipid metabolism of chickens.

The many studies have shown that probiotics could reduce cholesterol level in egg yolk [10, 11] and reduce cholesterol level in serum, carcass and liver of broiler chickens [8, 12, 13]. The study by Ramasamy showed that Lactobacillus culture in the feeding of hens can decrease the cholesterol level of eggs at 24 and 28 at 32 weeks of age [14]. Kimoto has been reported that a culture of *Lactobacillus acidophilus* taking up cholesterol actively by in vivo to exert a hypocholesterolemic effect [15].

It was known that some strains of *Bifidobacteria, Lactobacillus fermentum* KC5b, *Lactobacillus acidophilus johnsonii* and *Lactobacillus delbrueckii* JCM 1002 were able to remove cholesterol from culture medium during anaerobic condition growth in the presence of bile acid enrichment, and it depends on strains [16]. This research is important to prove the ability of lactic acid bacteria *Bifidobacterium sp* and *Lactobacillus acidophilus* to decrease cholesterol, LDL and increase HDL in the egg production.The objective of this recent study was to evaluate the effect of the combination probiotics contain *Bifidobacterium sp* and *Lactobacillus acidophilus* could affect on cholesterol, LDL and HDL in the egg yolk of laying hens.

2. Materials and methods

This research is quantitative. A total of 60 laying hens (Isa Brown) at 23 weeks of age was randomized under standard managed conditions. Each laying hen was placed into each battery cage (20x35x35 cm) of an open house system. The cages were assigned at random among 3 treatments, with 4 replicates, each with 5 laying hens; the treatments were as follows: 1) control, basal diet without probiotics and without AGPs; 2) basal diet with 0.1% AGPs; 3) basal diet with 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus*. Basal diets and water were provided ad libitum. At the end of 4-week period, eggs were selected at random from each treatment and the yolks separated. Duplicate sub-samples were taken and then analyzed for total cholesterol, LDL and HDL. The quantitative value of cholesterol, LDL and HDL were determined by using UV-Vis spectrophotometer methods.

All data treatment was analyzed using the analysis of variance procedure and treatment mean differences were tested using the least significant difference. The level of significance of the analyses was set at $p < 0.05$. The computation was done using the SPSS program 21 version.

3. Results and discussion

The effects of probiotics on the HDL level of egg yolk by using UV-Vis spectrophotometer presented in Table 1. The results of a recent study using probiotics showed that there was a significant difference ($p<0.05$) between the treatment of egg yolk HDL. The highest HDL level (50.00 mg/dL) was found in the treatment using 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus* and the lowest egg yolk cholesterol level was found in the control without probiotics and without AGPs (41.59).

Table 1. Average of high density lipoprotein (HDL) of egg yolk in the treatment.

Treatment	Average HDL (mg/dL)
P0=control	41.59 [a]
P1=0.1% AGPs	43.36 [b]
P2= 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus*	50.00 [c]

The effects of probiotics on the LDL level of egg yolk by using UV-Vis spectrophotometer presented in Table 2. The results of a recent study using probiotics showed that there was a significant difference ($p<0.05$) between the treatment of egg yolk LDL. The highest LDL level was found in the control without probiotics and without AGPs (58.63 mg/dL), while the lowest egg yolk cholesterol level was found in the treatment using 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus* (48.460 mg/dL).

Table 2. Average of low density lipoprotein (LDL) of egg yolk in the treatment.

Treatment	Average LDL (mg/dL)
P1=0.1% AGPs	48.463 [a]
P2= 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus*	48.460 [a]
P0=control	58.63 [b]

The effects of probiotics on cholesterol level of egg yolk by using UV-Vis spectrophotometer presented in Table 3. The effects of supplementation with *Bifidobacterium sp* and *Lactobacillus acidophilus* probiotic on egg yolk in laying hens showed there were significant differences between the treatment. The egg yolk cholesterol level was decreased ($p<0.05$) compared the control and AGPs treatment. The highest cholesterol level was found in the control without probiotics and without AGPs (224.02 mg/dL), while the lowest egg yolk cholesterol level was found in the treatment using 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus* (155.86 mg/dL).

Table 3. Average of cholesterol of egg yolk in the treatment.

Treatment	Average cholesterol (mg/dL)
P2= 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus*	155.86 [a]
P1=0.1% AGP	211.41 [b]
P0=control	224.02 [c]

This recent study in line with the other research that showed significant results in decrease of egg yolk cholesterol content by used the 0.1% Lactic Acid Bacteria, contain *Lactobacillus paracasei* (KKP 824), *Lactobacillus rhamnosus* (KKP 825) and *Lactobacillus rhamnosus* (KKP 826) (17) with concentration 6.7 $x10^8$ cfu/g [17]. The other research showed that the addition of probiotic had a significant effect on decrease egg yolk cholesterol (mg/g of yolk) [4]. The study by Haddadin showed that he giving of probiotic *Lactobacillus acidophilus* can decrease the egg cholesterol level in 40 weeks of production [10]. The recent study also in line with the study by Abdulrahim, the result showed that the effect of *Lactobacillus acidophilus* was a significant decrease in the cholesterol level in the egg yolks (16.95%) and serum (56.24%). Giving *Lactobacillus acidophilus* after 12 weeks of culture administration can reduce cholesterol level in the blood, through process deconjugating bile salts in the intestine, thereby preventing them from acting as precursors in cholesterol synthesis [18].

The use of Bifidobacteria sp in this recent study in line with a Liong's research show that Bifidobacteria strains could remove of cholesterol from the nutrient medium incorporated with cholesterol. The cholesterol removal was associated with growth of organisms. It was determined by the difference in cholesterol level in the medium after and before of the incubation period. Several mechanisms have been proposed to reduce cholesterol that included absorption cholesterol by growing cells of the intestine [16], cholesterol binding to cell surface [19, 20], incorporation into cellular membrane [15], deposition of cholesterol by conjugated bile salts [21].

The decreased absorption of cholesterol in the intestinal through co-precipitation of intestinal cholesterol with the deconjugated bile salt, which is observed at pH values below 5.5 [16]. However, some of these mechanisms depend on the strain bacteria and laboratory conditions [20]. It was

reported that fed broilers by probiotic (*Lactobacillus acidophilus, Bifidiobacteria, Aspergillus oryzae*) have lower levels of cholesterol compared with control group [22]. The decrease of cholesterol level by probiotic bacteria is due to the modulation of lipid metabolism, incorporation and assimilation of cholesterol in the cell membrane of the probiotics, intestinal conversion of cholesterol in coprostanol, and inhibition of the expression of the intestinal cholesterol transporter Niemann–Pick C1 like 1 (NPC1L1) in the enterocytes [23].

4. Conclusion

Based on the result, it could conclude that the supplementation of a combination 0.5% *Bifidobacterium sp* + 0.5% *Lactobacillus acidophilus* probiotic could improve egg yolk quality of laying hens by decreased cholesterol and LDL and increase HDL in egg yolk.

5. References

[1] Fuller R 1999 *Probiotics for farm animals, in Probiotics: A Critical Review*, (UK: Wymondham Horizon Scientific Press) pp 15-22
[2] Heak C, Sukon P and Sornplang P 2018 *Asian Australia. J. Anim.* **31,** 17-81
[3] Tannock G W 1999 *J Antimicrob Chemother* **43,** 849-852
[4] Tortuero F and Fernandez E 1995 *Anim. Feed. Sci. Technol* **53**, 255-265
[5] Balevi T, Ucan U S, Coskun B, Kurtoglu V and Cetingul I S 2001 *Br. Poult. Sci.* **42**, 456-461
[6] Davis G S and Anderson K E 2002 *Poult. Sci.* **81**, 755-759
[7] Ishihara N, Chu D C, Akachi S and Juneja L R 2000 *Poult. Sci.* **79**, 689-697
[8] Jin L Z, Ho Y W, Abdullah N and Jalaludin S 1998 *Poult. Sci.* **77,** 1259-1265
[9] Cisek A A and Binek M 2014 *Pol. J. Vet. Sci.* **17,** 385-389
[10] Haddadin M S Y, Abdulrahim S M, Hashlamoun E A R and Robinson R K 1996 *Poult. Sci.* **75**, 491-494
[11] Xu C L, Ji C, Ma Q, Hao K, Jin Z Y and Ki K 2006 *Poult. Sci.* **85**, 364-368
[12] Kalavathy R, Abdullah N, Jalaludin S and Ho Y W 2003 *Br. Poult. Sci.* **44**, 139-144
[13] Kalavathy R, Abdullah N, Jalaludin S, Wong C M V L and Ho Y W 2006 *Anim. Res.* **55,** 77-82
[14] Ramasamy K, Abdullah N, Jalaludin S, Wong M and Ho Y W 2009 *J. Sci. Food Agric.* **89**, 482-486
[15] Kimoto H, Ohmomo S and Okamoto T 2002 *J. Dairy. Sci* **85**, 3182-3188
[16] Pereira D I and Gibson G R 2002 *Appl. Environ. Microbiol.* **68,** 4689-4693
[17] Saleh A A, Gálik B, Arpášová H, Capcarová M, Kalafová A, Šimko M, Juráček M, Rolinec M, Bíro D and Abudabos A M 2017 *Ital. J. Anim. Sci.* **16**, 132-139
[18] Abdulrahim S M, Haddadin M S, Hashlamoun E A and Robinson R K 1996 *Br. Poult. Sci.* **37**, 341-346
[19] Noh D O, Kim S H and Gilliland S E 1997 *J. Dairy. Sci.* **80,** 3107-3113
[20] Liong M T and Shah N P 2005 *J. Dairy. Sci.* **88**, 55-66
[21] Marlett J A 1997 *Sites and Mechanisms for the Hypocholesterolemic Actions of Soluble Dietary Fiber Sources. In Dietary Fiber in Health and Disease* (Boston: Springer M A) pp 109-121
[22] Panda A K, Reddy M R, Rao S R, Raju M V and Praharaj N K 2000 *Archiv für Geflügelkunde* **64**, 152-156
[23] Reis S A, Conceição L L, Rosa D D, Siqueira N P and Peluzio M C 2017 *Nutr. Res. Rev.* **30**, 36-49.

Acknowledgement

The research team is grateful to the Ministry of Research and Technology of Higher Education (*Kemenristekdikti*), Chair of the Research and Innovation Institute, Rector Universitas Airlangga, Dean of Faculty of Veterinary Medicine Universitas Airlangga, for funding this research (PTUPT 2019).

Environmental Earth Sciences

Country	Germany -
Subject Area and Category	Agricultural and Biological Sciences Soil Science Earth and Planetary Sciences Earth-Surface Processes Geology Environmental Science Environmental Chemistry Global and Planetary Change Pollution Water Science and Technology
Publisher	Springer Verlag
Publication type	Journals
ISSN	18666299, 18666280
Coverage	2009-2020
Scope	Environmental Earth Sciences is an international multidisciplinary journal concerned with aspects of interaction between humans, natural resources or unique geographic zones, and the solid earth: -Ground water and soil contamination caused by waste management and disposal practices -Environmental problems associated with transportation by land or water -Geological processes that may impact ecosystems or humans -Man-made or naturally occurring geo-hazards -Environmental problems caused by extraction of minerals, coal, and ores, as well as oil and gas, water and alternative energy sources -Environmental impacts of exploration, remediation, and hazardous materials -Management of geoenvironmental data and information systems -Dissemination of knowledge on techniques, methods, approaches and experiences to improve the environment In pursuit of these topics, the geoscientific disciplines are invited to contribute their knowledge and experience. Major disciplines include: hydrogeology, hydrochemistry, geochemistry, geophysics, engineering geology, natural resources management, environmental geography and soil science.

107

H Index

Homepage

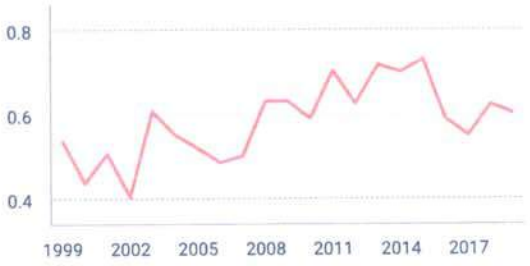
How to publish in this journal

Contact

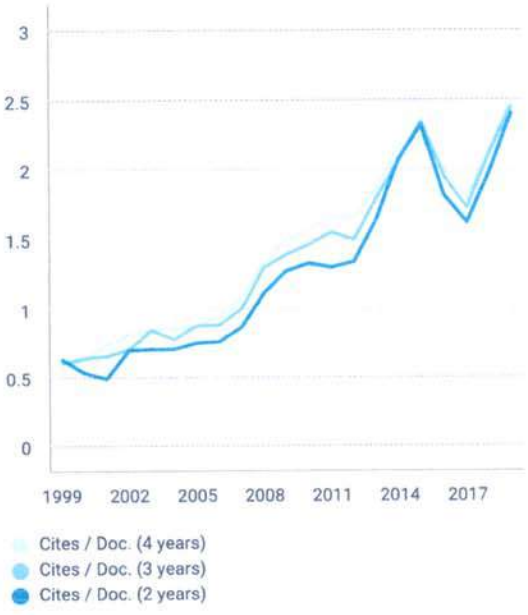
Join the conversation about this journal

Quartiles

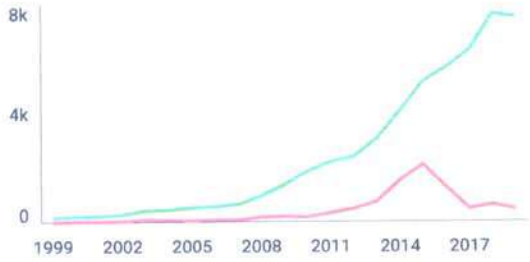
SJR



Citations per document

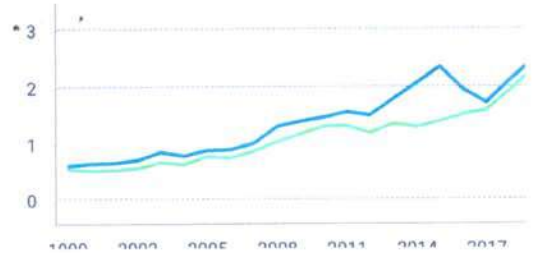


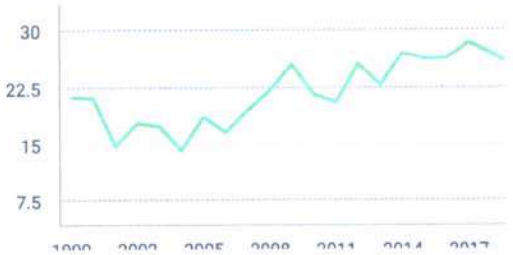
Total Cites Self-Cites



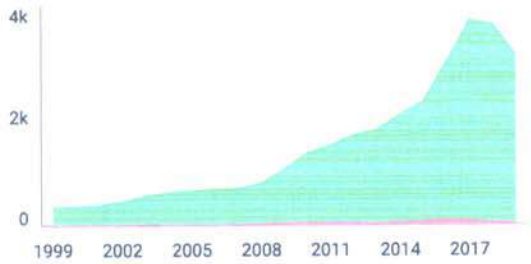
External Cites per Doc Cites per Doc

% International Collaboration

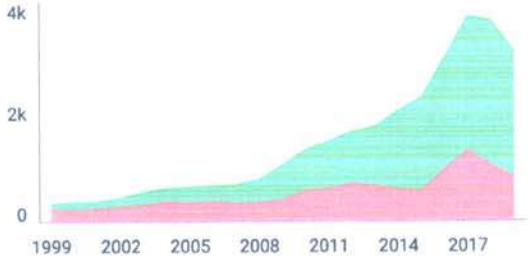




Citable documents Non-citable documents



Cited documents Uncited documents



← Show this widget in your own website

Just copy the code below and paste within your html code:

<a href="https://www.scima

Climate Transparency Report

A comprehensive evaluation on how Indonesia doing in handling climate change

iesr.or.id

Metrics based on Scopus® data as of April 2020

Adept 6 months ago

Hi, do you rank the journals (quartiles) based on the amount citations per year?

And do the rankings (quartiles) here determine the level of prestige (of the journals), in the eyes of all academic institutions?