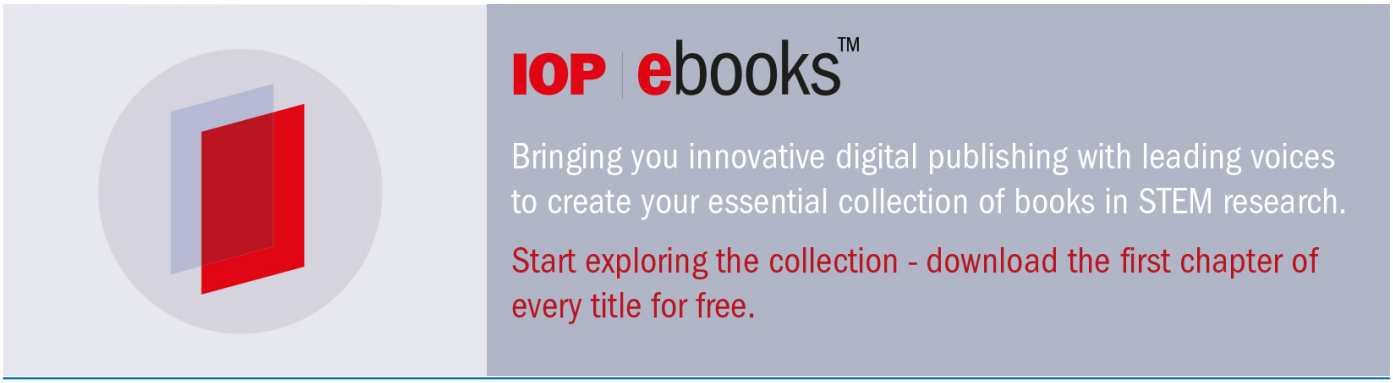
Characterization of thermostable chitinase from *Bacillus licheniformis* B2



IOP Conf. Series: Earth and Environmental Science **293** (2019) 012030 doi:10.1088/1755-1315/293/1/012030

Characterization of thermostable chitinase from *Bacillus licheniformis* B2

N Jayanthi, M G M Purwanto*, R Chrisnasari, T Pantjajani, A Wahjudi and M Sugiarto

Department of Biology, Faculty of Biotechnology, University of Surabaya, Jl. Raya Kalirungkut, Surabaya 60292, Indonesia

*Corresponding author: maria_gmp@staff.ubaya.ac.id

Abstract. Chitinases is an enzyme capable of degrading chitin into oligomers to produce chitin derivatives products which are more useful. Thermostable-chitinase is of important in the relevant industrial application, since the degradation process oftently requires prety high temperature. This research report a characterization of chitinase isolated from thermophilic microorganism. The chitinase was obtained from *Bacillus licheniformis* B2 isolated from Ijen hot spring, East Java. It has the best chitinolytic activity at pH 7 when colloidal chitin was used as substrate. The enzyme exhibited activity in broad temperature range, from 50 °C to 70 °C, optimally at 55 °C. It was stable at 50 °C up to 90 min, at 60 °C up to 60 min and at 70 °C up to 30 min. At neutral pH this enzyme has negative charge but further purification is needed to determine its pI. The K_m and V_{max} of this chitinase for colloidal chitin were 101.96 mg mL^{-1} and 2.72 μmol $(min\ mL)^{-1}$, respectively. Addition of NaCl, KNO_3 and $MgSO_4$ decreased the activity of chitinase following mixed inhibitor mode. This enzyme should be a good candidate for applications in the recycling of chitin waste.
Keywords: *Bacillus licheniformis*, chitinase, shellfish waste, waste to food

1. Introduction

Chitinase (EC 3.2.1.14) is a group of enzyme capable of degrade chitin to low-molecular-weight products. The production of inexpensive chitinolytic enzymes is an important element in the utilization of shellfish wastes that not only solves environmental problems but also promotes the economic value of marine products [1]. Chitinase can be isolated from many kind of organism, such as fungi, bacteria [2], plant and animal. Thermostable-chitinase usually produce by thermophilic microorganism at geothermal environment [3] for instance volcano area and hot springs.

Enzyme work optimally in specific condition, such as temperature, pH, substrate concentration and the presence of inhibitor and activator [4]. Those aspect affect three dimension comformation and polypeptide folding of enzyme. Ion addition to enzymatic reaction can enhance enzyme activity and work as activator or inhibit enzyme and work as inhibitor [5]. In this experiment, NaCl, KNO_3 and $MgSO_4$ used because of its soluble property in phosphate buffer. Many study has been done to characterize thermostable chitinase produced by microorganism but the role of each ion in enzyme activity varies depending on the type chitinase produced and the producing bacteria species.

Previously, *Bacillus licheniformis* B2 that produced a particular high activity of thermostable chitinase was obtained when cultured in the medium containing colloidal chitin as the sole carbon

source. According to BLAST analysis, this microorganism have the highest similarity with *B. licheniformis* strain PR with 95 % identification. Crude chitinase from this microorganism was successfully used to prepare low-molecular- weight chitin monomer and oligomers. This paper describes the characterization and some properties of chitinases from the culture medium of *B.licheniformis* B2.

2. Materials and methods

2.1. Materials

Bacillus licheniformis B2 isolated from Ijen hot spring, chitin extracted from shrimp shell, KNO_3 (Merck), $MgSO_4$ (Merck), HCl (J. T. Baker), NaOH (Merck), $CaCl_2$ (Merck), $(NH_4)_2SO_4$ (Merck), K_2HPO_4, NaCl (Merck), $MgSO_4.7H_2O$, yeast extract (Becton & Dickinson), bacto tryptone (Merck), H_2O, Na_2HPO_4(Merck), NaH_2PO_4 (Merck), Na_2CO_3, Na_2SO_4, $CuSO_4$, H_2SO_4, ammonium molybdat, $NaHAsO_4.7H_2O$, BSA, phosphoric acid 85 % (Merck), acetic acid (Merck), CTAB, Bio-rex (Bio-Rad), DEAE cellulose (Sigma Aldrich), GlcNAc (Carbosynth).

2.2. Production of chitinase

Colony of B. *licheniformis* B2 inoculated to thermus broth without chitin and incubated at 50 °C with 180 rpm shaking for 12 h or until maximum density. Afterwards, to make starter add 7.5 mL of culure to 67.5 mL of thermus broth media and incubated for 16 h at 50 °C with 180 rpm shaking (1 rpm = 1/60 Hz). Total of 75 mL starter then added to fermentor with 675 mL thermus broth media. The production of chitinase is carried out with following condition, temperature 50 °C, pH 5, 200 rpm agitation and 2 L min^{-1} aeration. After incubation, all media at fermentor centrifuged at 11 000 rpm for 5 min to separate chitin and cell debris (pellet) with chitinase crude extract (supernatant).

2.3. Enzyme assay

Chitinase activity was assayed in a 0.5 mL reaction mixture containing 0.1 g mL^{-1} colloidal chitin in 0.02 M phosphate buffer, pH 7, and 0.5 mL of enzyme solution. After incubation at 55 °C for 90 min, the reaction was stopped by centrifugation. For every 10 min, the tube was shaken to maximize enzyme-substrate contact. Reducing sugar produced was measured by Nelson-Somogyi method. Chitinase activity was defined using equation (1) [6].

$$\text{Activity}\left(\frac{\mu\text{mol}}{\text{min mL}}\right) = \frac{\text{GlcNAc released per mL} \times \text{Total assay volume}}{\text{Mr GlcNAc} \times \text{Duration of incubation} \times \text{Volume of enzyme used}} \quad (1)$$

Notes:
GlcNAc released per mL = GlcNAc mass per mL (μg mL^{-1})
Mr GlcNac = 221.21 (g mol^{-1})

2.4. Optimum temperature and pH

The optimum temperature and pH were measured using colloidal chitin as a substrate. The enzyme activity was assayed for 90 min at temperatures 40 °C to 65 °C and pH 4.0 to 8.0 using 0.02 M phosphate buffer.

2.5. Thermal stability

The thermal stability was investigated by incubating the enzyme for 2 h at temperatures 50 °C, 60 °C, 70 °C, 80 °C, and 90 °C in phosphate buffer pH 7.0. Aliquots were taken every 30 min, and residual activity of the enzyme was determined under standard assay condition.

2.6. pI determination
To determine enzyme's pI, buffer pH 4 to 9 was used with addition of five drops 0.1 % CTAB (cationic wetting agent) for each mL of buffer. Buffer with CTAB was then added to 0.2 mL of enzyme with different pH at separate tube then turbidity of each tube was observed visualy.

2.7. Ion exchange chromatography
Column resin was prepared corresponding to each protocol, i.e. bio-rex resin for cation exchange chromatography and DEAE cellulose resin for anion exchange chromatography. Resin was to column and flow rate was set to 1 mL min^{-1}. The amount of 2 mL enzyme was loaded then eluted using phosphate buffer pH 7 with gradient concentration of NaCl. Protein concentration was determined using Bradford method in every fraction obtained from chromatography.

2.8. Molecular weight determination
Separating gel (12.5 %) was prepared with the composition as follows: 3.125 mL acrylamide 30 % (29.2 g acrylamide and 0.8 g bis-acrylamide in 100 mL aquades); 2.75 mL Tris-HCl 1 M pH 8.8; 1.505 mL aquadest; 75 µL SDS 10 %; 75 µL APS 10 % and 6.25 µL TEMED. Stacking gel was prepared on top of separating gel with the composition as follows: 0.45 mL bis-acrylamide 30 %; 0.38 mL Tris-HCl 1 M pH 6.8; 2.11 mL aquabidest; 30 µL SDS 10 %; 5 µL TEMED and 30 µL APS 10 %. Sample was prepared by adding sample buffer and boiled for 5 min. Total of 10 µL to 20 µL of sample was then loaded into the well. After electrophoresis run for 20 min to 30 min with 110 mA, the gel was stained with staining solution (1 g Coomassie Brilliant Blue G-250, 450 mL methanol, 450 mL aquadest and 100 mL acetic acid) and then destained with destaining solution (100 mL methanol, 100 mL acetic acid and 800 mL aquadest). Protein bands shown from sample were then compared to protein marker to determine enzyme molecular weight.

2.9. Effect of salt addition
To determine effect of salt addition, following variation was added to enzyme assay: 0.47 mM and 1.3 mM NaCl, 0.47 mM and 1.3 mM KNO_3, 0.47 mM and 0.238 mM $MgSO_4$. Enzyme activity was tested in 7 different substrate concentration (2.5 mg mL^{-1} to 17.5 mg mL^{-1} colloidal chitin) and compared to the activity without any salt addition.

3. Results

3.1. Temperature and pH characterization
The enzyme showed optimal activity at pH 7 (figure 1). At pH 7, the enzyme had the highest chitinolytic activity at 55 °C (figure 2). The half-life at 50 °C was about 90 min, whereas at 60 °C and 70 °C, no significant activity lost was observed within 60 min and 30 min, respectively (figure 3).

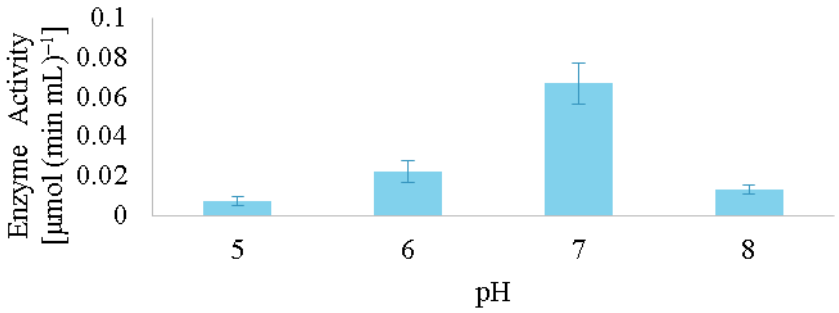


Figure 1. Optimum pH of chitinase from *B. licheniformis* B2.

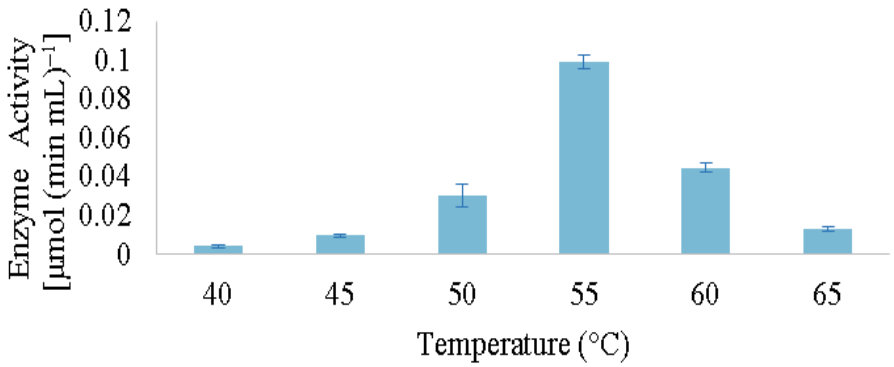


Figure 2. Optimum temperature of Chitinase from *B. licheniformis* B2.

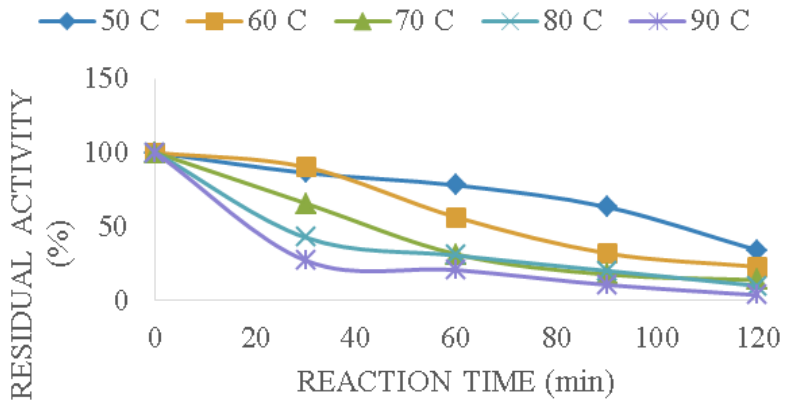


Figure 3. The thermal stability of chitinase from *B. licheniformis* B2.

3.2. Protein charge determination

The result of pI determination using CTAB can be observed at figure 4. All solution gave almost same turbidity in pH 4 to pH 10 and enzyme pI can't be determined. As shown at the graph (figure 5 and figure 6), enzyme can't attach to column with negative charge and can be detect at early fraction beside at column with positive charge, high concentration of salt is needed to unattached enzyme from column so enzyme present in later fraction.

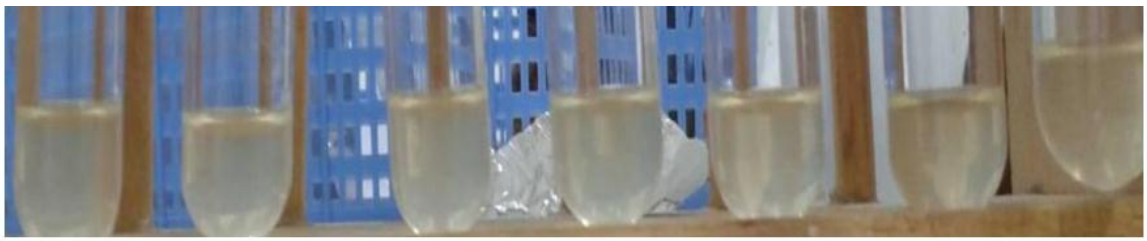


Figure 4. Enzyme at buffer with 0.1 % CTAB, from left to right: pH 4, 5, 6, 7, 8, 9, and 10.

3.3. Ion exchange chromatography

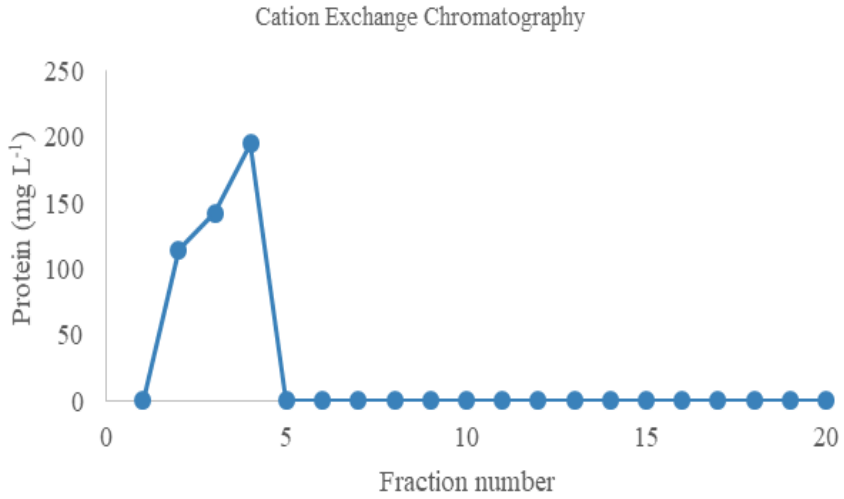


Figure 5. Protein concentration in each fraction after IEC (1 mL per fraction)

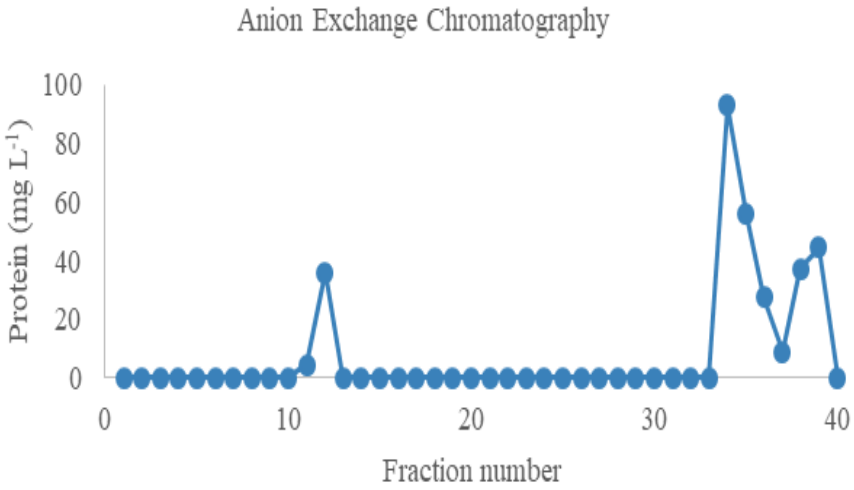


Figure 6. Protein concentration in each fraction after IEC (0.5 mL per fraction)

3.4. Molecular weight determination

Enzyme in crude extract visualized using SDS PAGE method with CBBG R250 staining. Visualization done to determine enzyme molecular weight. As shown in figure 7, in crude extract from B. *licheniformis* there were several protein bands shown. Proteins contained in crude extract sized between 50 kDa and 75 kDa, between 25 kDa and 35 kDa, between 15 kDa and 25 kDa, and also between 10 kDa and 15 kDa. It remains unclear which protein has chitinolytic activity.

3.5. Enzyme kinetic

The kinetic of the enzyme was studied on colloidal chitin. The kinetics of chitinase from *B. licheniformis* B2 followed the classical Michaelis–Menten kinetics. The Km and Vmax value calculated from Lineweaver–Burk plots was 101.96 mg mL^{-1} and 2.72 µmol $(min\ mL)^{-1}$, respectively (table 1). Addition of NaCl (figure 7), KNO_3 (figure 8), and $MgSO_4$ (figure 9) reduced both Km and Vmax. Increasing salt concentration also increasing Km and Vmax impairment.

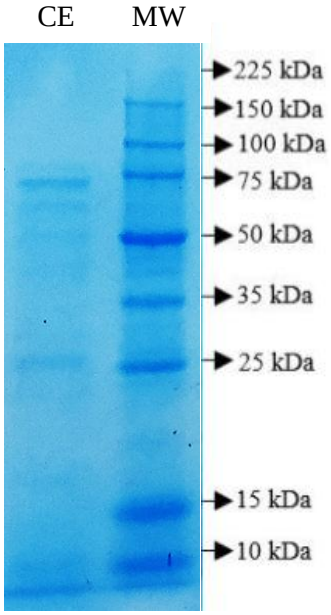


Figure 7. Protein in crude extract (CE) visualization using SDS PAGE method.

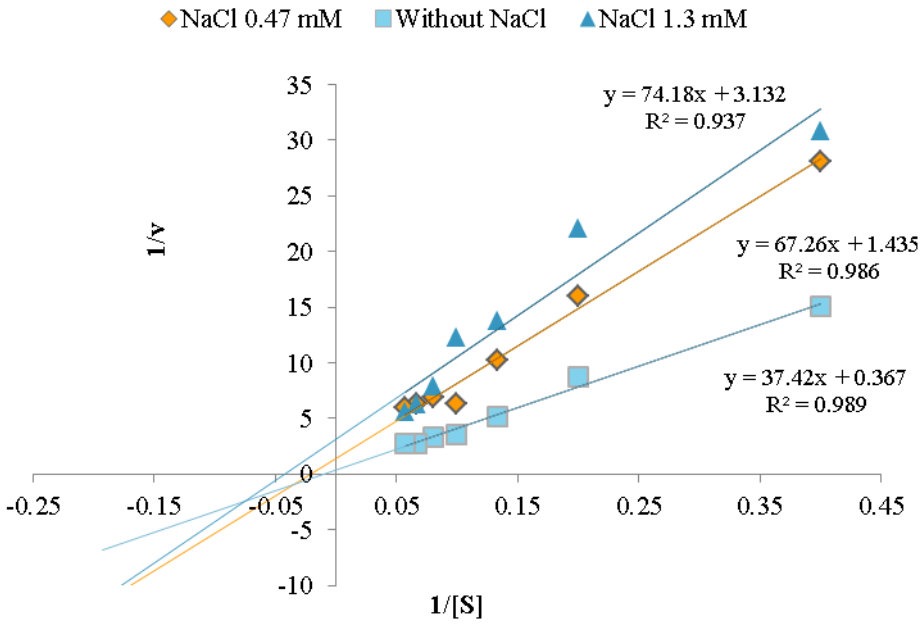


Figure 7. Lineweaver-Burk Plot of Chitinase with NaCl addition

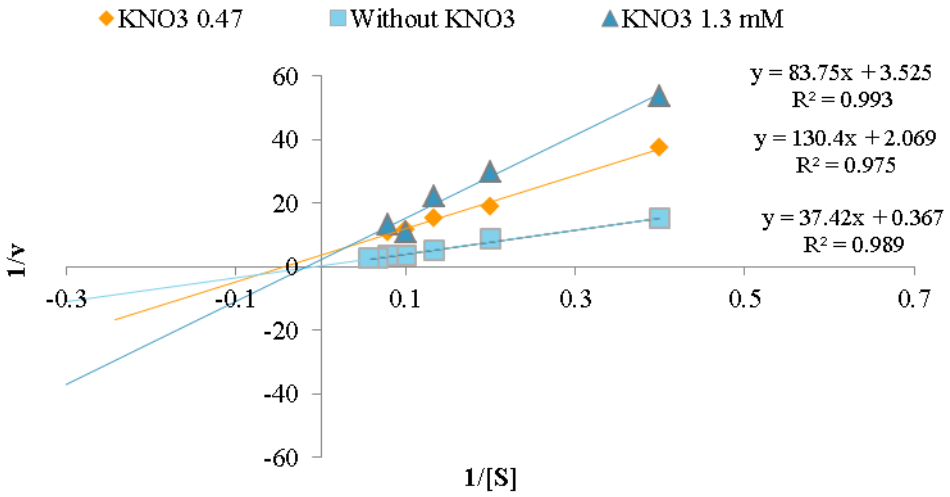


Figure 8. Lineweaver-Burk Plot of Chitinase with KNO_3 addition.

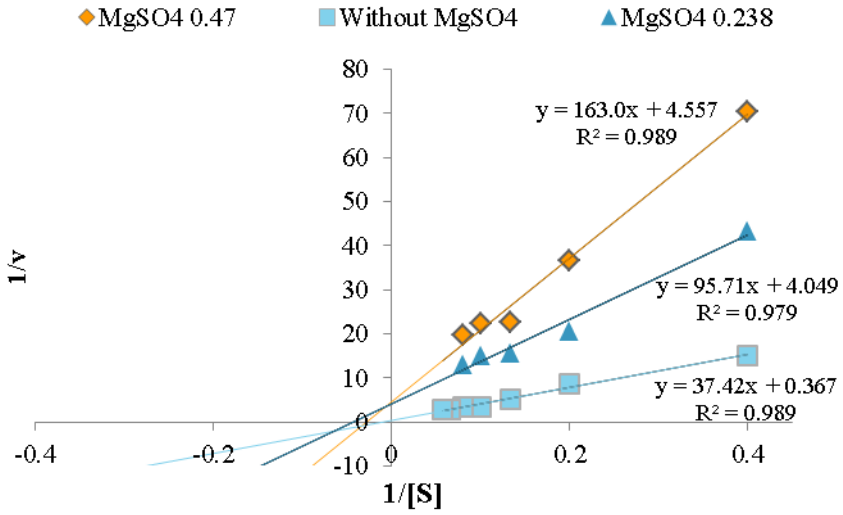


Figure 9. Lineweaver-Burk Plot of Chitinase with $MgSO_4$ addition.

Table 1. Km and Vmax value with salt addition.

	Km (mg mL^{-1})	Vmax (µmol $(min\ mL)^{-1}$)
Without salt	101.96	2.72
0.47 mM NaCl	46.87	0.69
1.3 mM NaCl	23.68	0.31
0.47 mM KNO3	63.02	0.48
1.3 mM KNO3	23.76	0.28
0.47 mM MgSO4	35.76	0.21
0.238 mM MgSO4	23.63	0.24

4. Discussion

B. licheniformis B2 was isolated from Ijen hot spring, East Java, Indonesia. Temperature and pH characterization of chitinase obtained from this microorganism were carried out to know the best condition when using chitinase to degrade shrimp shell waste. Variation of pH 5 to pH 8 was chosen based on sampling environment pH, which is 6. Variation of temperature was also taken based on sampling environment temperature, which is 50 °C. The enzyme showed optimal activity at pH 7 with average activity 6.69×10^{-2} µmol (min mL)$^{-1}$ (figure 1). Activity at pH 7 was the highest and significantly different compared to activity at other pH when analyzed with One Way ANOVA (data not shown). At pH 7, the enzyme had the highest chitinolytic activity at 55 °C with average activity 9.95×10^{-2} µmol (min mL)$^{-1}$ (figure 2). Activity at 55 °C also significantly different compared to activity at other pH when analyzed with One Way ANOVA (data not shown).

The thermostability of enzyme was observed based on residual activity when enzyme was heated at certain temperature or enzyme half-time. According to the result, at 50 °C the residual activity remained 63.24 % after 90 min and decreased to 33.98 % after 120 min, at 60 °C the residual activity remained 56.34 % after 60 min and 31.99 % after 90 min, at 70 °C after 30 min the residual activity remained 65.72 % after 30 min and continued to decline afterwards. The enzyme is called stable when its residual activity is greater than 50 %. It was stable at 50 °C up to 90 min, at 60 °C up to 60 min and at 70 °C up to 30 min. While at 80 °C and 90 °C the enzyme was no longer stable.

An trial experiment was also done to determine pI using ionic detergent, i.e. CTAB. In buffer with same pH as enzyme pI, enzyme should have neutral charge and cause high turbidity. As shown in figure 4, buffer with pH 4 to pH 10 gave almost the same turbidity so pI enzyme could not be determined. This phenomenon can be happen because enzyme crude extract contained more than one protein with different pI and further purification is needed to determine the exact pI of this chitinase. Enzyme showed different characteristics when separated in positively charge column compared to that in negatively charge column. As shown in figure 5, chitinase was obtained at early fraction. This result proofed that the enzyme has similar charge with column used (negative) or on its net-charge at pH 7. In figure 6, protein presented at early and later fraction. This result shown that high concentration salt needed to unattached this protein from the column and chitinase has different charge with column used (positive). Combined these two results, it was concluded that chitinase from *B. licheniformis* B2 had negative charge at pH 7.

Addition of 0.47 mM and 1.3 mM NaCl caused depletion of activity up to 46 % and 63 % respectively. Addition of KNO_3 with the same concentration also inhibited enzyme activity. Addition of 0.47 mM KNO_3 decrease 60 % of enzyme activity and addition of 1.3 mM KNO_3 decrease 71.7 % of enzyme activity. Addition of 0.47 mM and 0.238 mM of $MgSO_4$ also caused inhibition of enzyme. This result obtained because addition of salt increased the amount of ions in solution. Those ions can interact with enzyme or substrat-enzyme complex and decreasing product-making reaction. From these result we can conclude that NaCl, KNO_3 and $MgSO_4$ act as inhibitor for chitinase from *B. licheniformis* B2. The result was similar to characterization of chitinase from T5a1 [7], addition of 1 mM Mg^{2+}, Na^{+} and K^{+} caused enzyme activity depletion. Different result obtained at other species, NaCl can act as activator for chitinase from *Enterobacter* sp. G-1 [8].

Enzyme Km and Vmax was obtained by enzyme assay with various substrate concentrations. The reaction was held for 90 min at 55 °C. The enzyme followed the classical Michaelis–Menten kinetics so the amount of Km and Vmax can be determined using Lineweaver-Burk plot. Chitinase from *B. licheniformis* B2 showed Km and Vmax value 101.96 mg mL^{-1} and 2.72 µmol (min mL)$^{-1}$, respectively. Different enzyme will give different Km and Vmax, Km and Vmax chitinase from *B. licheniformis* JP2 were 0.321 µg mL^{-1} and 71.429 µg, while Km and Vmax chitinase from *Enterobacter* sp. NRG4 were 1.41 mg mL^{-1} and 74.07 µM (µg h)$^{-1}$, respectively [9]. Km and Vmax chitinase from *Serratia marcescens* B4A was 8.3 mg mL^{-1} and 2.4 mmol min^{-1} [10], Km and Vmax chitinase from B. *cereus* 11 UJ were 29.71 µg mL^{-1} and 1.035×10^{-1} µg (mL s)$^{-1}$, respectively [11]. The lower the Km, the higher the enzyme activity towards colloidal chitin as substrat. Vmax or maximum velocity of this enzyme was 2.72 µmol (min mL)$^{-1}$ which mean at optimum condition,

enzyme can converte colloidal chitin to 2.72 µmol mL^{-1} GlcNAc each minute. If both Vmax and Km value decreased and slope at Lineweaver Burk plot changed after addition of inhibitor, that inhibitor inhibites enzyme with mixed type. Mixed type inhibitor is inhibitor that binds to allosteric site of enzyme and not the active site of enzyme. Mixed inhibition can caused increament of enzyme affinity toward substrate but did not increase product formation [12].

5. Conclusion

Chitinase as a good candidate for applications in the recycling of chitin waste was obtained from *B. licheniformis* B2 isolated from Ijen hot spring, East Java. It showed best chitinolytic activity at pH 7 and exhibited activity in broad temperature range, from 50 °C to 70 °C, optimally at 55 °C. The Km and Vmax of this chitinase for colloidal chitin were 101.96 mg mL-1 and 2.72 µmol $(min\ mL)^{-1}$, respectively.

Acknowledgments

The research work was supported by the Stranas Research Grant 004/SP2H/K7/KM/2018 form DIKTI (Directorate General of Higher Education), Indonesia.

References

[1] Bhagat J, Venkatramani M, Hussain A J and Jayaprakashvel M 2014 Deproteination of shrimp shell wastes using immobilized marine associated Pseudomonad Amet1776 *Biosciences Biotechnology Research Asia* **11**(1) 211–20
https://www.researchgate.net/publication/267866886_Deproteination_of_Shrimp_Shell_Wastes_using_Immobilized_Marine_Associated_Pseudomonad_Amet1776

[2] Huang C J and Chen C Y 2004 Gene Cloning and biochemical characterization of chitinase ch from *Bacillus Cereus* 28-9 *Ann. Microbiol.* **54**(3) 289–97
https://www.researchgate.net/publication/228719243_Gene_cloning_and_biochemical_characterization_of_chitinase_CH_from_Bacillus_cereus_28-9

[3] Van der Burg B 2003 Extremophiles as a source for novel enzymes *Curr. Opin. Microbiol.* **6**(3) 213–18
https://www.ncbi.nlm.nih.gov/pubmed/12831896

[4] Robinson P K 2015 Enzymes: principles and biotechnological applications *Essays Biochem.* **15** (59) 1–41
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4692135/

[5] Murrsy R K 2014 *Biokimia Harper* [Harper Biochemistry] (Jakarta: ECG) [in Bahasa Indonesia]
https://www.belbuk.com/biokimia-harper-edisi-29-p-3870.html

[6] Chakraborty A and Mahajan A 2014 Cellulase activity enhancement of bacteria isolated from oil-pump soil using substrate and medium optimization Am. J. Microbiol. Res. Vol 2 (2) 52–6
http://pubs.sciepub.com/ajmr/2/2/1/index.html

[7] Noviendri D, Fawzya Y N and Chasanah E 2008 Karakteristik dan sifat kinetika enzim kitinase dari isolat bakteri T5a1 asal terasi [Characteristics and kinetics of chitinase enzyme from T5a1 bacterial isolates from shrimp paste] *Jurnal Pascapanen dan Bioteknologi Kelautan dan Perikanan* **3** No. 2 123–29 [Bahasa Indonesia]
http://www.bbp4b.litbang.kkp.go.id/jurnal-jpbkp/index.php/jpbkp/article/view/15

[8] Park J K, Morita K, Fukumoto I, Yamasaki Y, Nakagawa T, Kawamukai M and Matsuda, H 1997 Purification and characterization of the chitinase (ChiA) from *Enterobacter* sp. G-1. *Bioscience Biotechnology and Biochemistry* **61**(4) 684–89
https://www.tandfonline.com/doi/abs/10.1271/bbb.61.684

[9] Dahiya N, Tewari R, Tiwari R P and Hoondal G S 2005 Chitinase from *Enterobacter* sp. NRG4: Its purification, characterization and reaction pattern *Electron. J. Biotechnol.* **8**(2) 134–45
http://www.ejbiotechnology.info/index.php/ejbiotechnology/article/view/v8n2-6/448
[10] Zarei M, Aminzadeh S, Zolgharnaen H, Shafahieh A, Ghoroghi A, Motallebi A and Lotfi A S 2010 *Serratia marcescens* B4A chitinase product optimization using Taguchi approach *Iran. J. Biotechnol.* 8(4) 252–62
http://www.ijbiotech.com/article_7099_229a09fc874016b1155794ffc98e3bb4.pdf
[11] Suryadi Y, Priyatno T P, Susilowati D N, Samudra I M , Yudhistira N and Purwakusumah E D 2013 Isolasi dan karakterisasi kitinase asal *Bacillus cereus* 11 UJ [Isolation and Characterization of Chitinase from Bacillus cereus 11 UJ] *Jurnal Biologi Indonesia* 9(**1**) 51–62 [in Bahasa Indonesia]
https://media.neliti.com/media/publications/77635-ID-isolasi-dan-karakterisasi-kitinase-asal.pdf
[12] Storey K B 2004 *Functional Metabolism: Regulation and Adaptation* (New Jersey: Wiley)
https://www.wiley.com/en-us/Functional+Metabolism%3A+Regulation+and+Adaptation-p-9780471410904

ISSN 1755-1307

IOP Conference Series
Earth and Environmental Science

1st Workshop on Metrology for Agriculture and Forestry (METROAGRIFOR)

275

VOLUME 275 – 2019

3–5 October 2018
Ancona, Italy

EDITOR
Enrico Primo Tomasini

The open access journal for conference proceedings

iopscience.org/jpcs

IOP Publishing

IOP Conference Series: Earth and Environmental Science

The open access *IOP Conference Series: Earth and Environmental Science (EES)* provides a fast, versatile and cost-effective proceedings publication service.

RSS Sign up for new issue notifications

IOP Conference Series: Earth and Environmental Science

COUNTRY

United Kingdom

Universities and research institutions in United Kingdom

SUBJECT AREA AND CATEGORY

Earth and Planetary Sciences
Earth and Planetary Sciences (miscellaneous)

Environmental Science
Environmental Science (miscellaneous)

PUBLISHER

IOP Publishing Ltd.

H-INDEX

26

PUBLICATION TYPE

Conferences and Proceedings

ISSN

17551307, 17551315

COVERAGE

2010-2020

INFORMATION

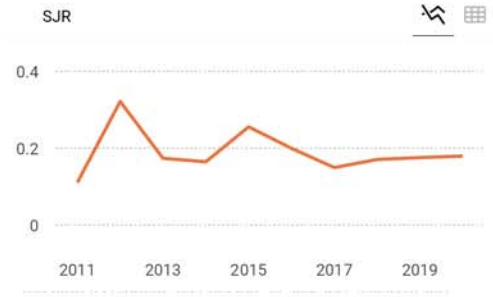
Homepage

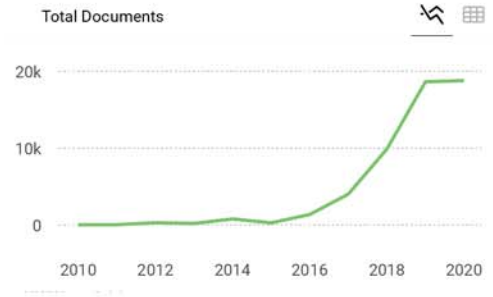
How to publish in this journal

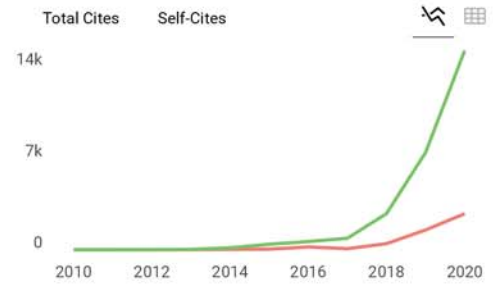
ees@ioppublishing.org

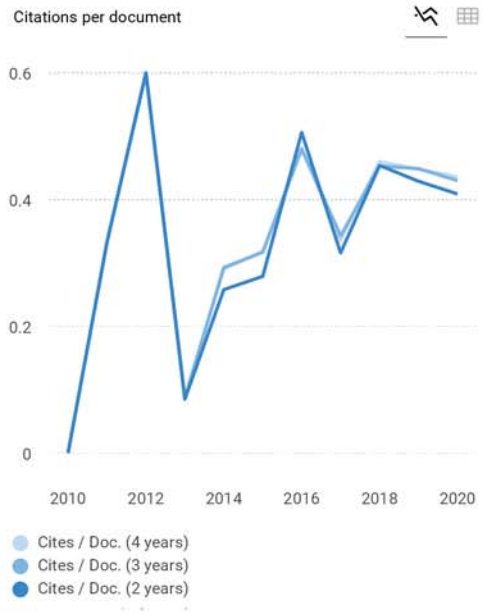
SCOPE

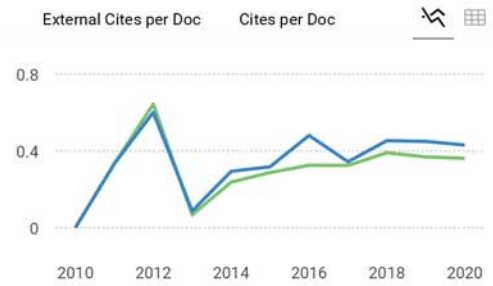
The open access IOP Conference Series: Earth and Environmental Science (EES) provides a fast, versatile and cost-effective proceedings publication service.

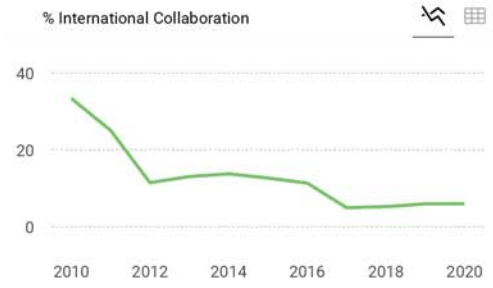


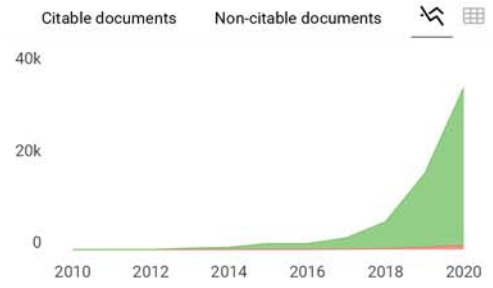


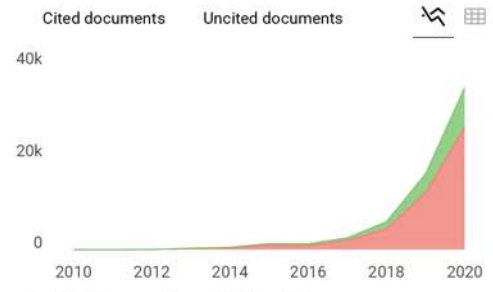












← Show this widget in your own website

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

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

Metrics based on Scopus® data as of April 2021

A **Alharia Dinata** 7 months ago

IOP Conference Series: Earth and Environmental Science - Volume 708 is not available in Scopus.

reply

SCImago Team

Melanie Ortiz 7 months ago

Dear Alharia,
thank you very much for your comment, unfortunately we cannot help you with your request. We suggest you contact Scopus support: https://service.elsevier.com/app/answers/detail/a_id/14883/kw/scimago/supporthub/scopus/
Best Regards, SCImago Team

V **Vani** 7 months ago

good evening, whether this journal is Q4 or Q2 ?

reply

SCImago Team

Melanie Ortiz 7 months ago

Dear Vani,
Thank you for contacting us.
As said below, we calculate the SJR data for all the publication's types, but the Quartile's data are only calculated for Journals and Book Series.
Best regards, SCImago Team

F **FEROSKHAN M** 12 months ago

IOP Conference Series: Earth and Environmental Science - Volume 573 is not available in Scopus. But later volumes are available. May I know when will they publish in Scopus?

reply

SCImago Team

Melanie Ortiz 12 months ago

Dear Sir/Madam,

thank you very much for your comment, unfortunately we cannot help you with your request. We suggest you contact Scopus support: https://service.elsevier.com /app/answers/detail/a_id/14883/kw/scimago/supporthub/scopus/
Best Regards, SCImago Team

N **Natt** 1 year ago

I would like to know the quartile of this journal. Why isn't it showing on the website?

reply

SCImago Team

Melanie Ortiz 1 year ago

Dear Natt,
Thank you for contacting us. We calculate the SJR data for all the publication's types, but the Quartile's data are only calculated for Journals and Book Series.
Best regards, SCImago Team

N **Nurgustaana** 2 years ago

Dear SCImago Team!
I want to know previous quartiles of journal (for 2018 and 2019 years). I have tried find information about a quartile, but discovered just SJR for 2018. Could you please provide information about it?
Yours sincerely, Nurgustaana

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Nurgustaana,
Thank you for contacting us. We calculate the SJR data for all the publication types, but the Quartile data are only calculated for Journal type's publications. Best regards, SCImago Team

M **Mora** 2 years ago

hello, how to search one of journal who publised by IOP, because when i find it by the title, they are not able in scimagojr but the publisher is available in here, thank you for the respond it means a lot

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Mora,

thank you for contacting us. Could you provide us the Title of the journal? We remember that SCImago Journal & Country Rank shows all the information have been provided by Scopus. If you didn´t localize the journal in the search engine, it means that Scopus / Elsevier has not provided us the corresponding data.

Best Regards, SCImago Team

D **Dr. Yousif** 2 years ago

Dear Sir,

I have published a paper in Earth and Environmental Science Journal (only myself, single author) I am trying to withdraw it after 28 days of publishing online, is it possible? Could you please tell me the procedure of withdrawing a paper?
Thank you,

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Yousif,
thank you for contacting us.
We are sorry to tell you that SCImago Journal & Country Rank is not a journal. SJR is a portal with scientometric indicators of journals indexed in Elsevier/Scopus.
Unfortunately, we cannot help you with your request, we suggest you to contact the journal's editorial staff , so they could inform you more deeply. Best Regards, SCImago Team

A **Agustinus Kastanya** 2 years ago

need information about renking of the Journal on Scopus

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Agustinus, thank you very much for your comment, unfortunately we cannot help you with your request. We suggest you to consult the Scopus database directly. Remember that the SJR is a static image of a database (Scopus) which is changing every day. Best regards, SCImago Team

M **Mahipal** 2 years ago

Dear Admin,

How could our journal include in your IOP?

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Mahipal,
thank you for contacting us.
We suggest you to contact the IOP's editorial staff , so they could inform you more deeply.
If you would like to make an application to Scopus, please contact them to help you with this issue here: https://www.elsevier.com/solutions/scopus/content/content-policy-and-selection
http://suggestor.step.scopus.com/suggestTitle/step1.cfm
Best Regards, SCImago Team

M **Mursalin** 2 years ago

Dear SCImago Team

My name is mursalin from Jambi City, Indonesia. I have published my article titled The Effect of Temperature on MDAG Purification Using Creaming Demulsification Technique at the IOP Conference Series: Earth and Environmental Science, Volume 309, conference 1 and could be accessed at: https://iopscience.iop.org/article/10.1088/1755-1315/309/1/012068. But why until now it does not appear into Google Scholar and my account.

Please help me to resolve the issue. Thank you for your kindness. I am waiting for good news from you.

Sincerelly,
Mursalin

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear Mursalin,

Thank you for contacting us. Unfortunately, we can not help you with your request. Maybe other users can help you. Best Regards, SCImago Team

N **Танзиля Созаева** 2 years ago

Доброго времени суток! Интересует информация по квартилю журнала (издания)

reply

SCImago Team

Melanie Ortiz 2 years ago

Dear user, thank you very much for your request. You can consult that information in SJR website. Best Regards, SCImago Team

S **syaiful** 3 years ago

I am very interested to send my paper to this conference

best regards

syaiful

reply

SCImago Team

Elena Corera 3 years ago

Dear user, in the link below you will find the information corresponding to the author's instructions of this journal. Best regards, SCImago Team
https://publishingsupport.iopscience.iop.org/author-guidelines-for-conference-proceedings/

Leave a comment

Name

Email
(will not be published)

I'm not a robot
reCAPTCHA
Privacy - Terms

Submit

The users of Scimago Journal & Country Rank have the possibility to dialogue through comments linked to a specific journal. The purpose is to have a forum in which general doubts about the processes of publication in the journal, experiences and other issues derived from the publication of papers are resolved. For topics on particular articles, maintain the dialogue through the usual channels with your editor.

Developed by: SCImago

Powered by: Scopus

Follow us on @ScimagoJR

EST MODUS IN REBUS
Horatio (Satire 1,1,106)

Source details

IOP Conference Series: Earth and Environmental Science

Scopus coverage years: from 2010 to Present

ISSN: 1755-1307 E-ISSN: 1755-1315

Subject area: Environmental Science: General Environmental Science
Earth and Planetary Sciences: General Earth and Planetary Sciences
Physics and Astronomy: General Physics and Astronomy

Source type: Conference Proceeding

CiteScore 2020
0.5

SJR 2020
0.179

SNIP 2020
0.436

View all documents > Set document alert Save to source list Source Homepage

CiteScore CiteScore rank & trend Scopus content coverage

Improved CiteScore methodology

CiteScore 2020 counts the citations received in 2017-2020 to articles, reviews, conference papers, book chapters and data papers published in 2017-2020, and divides this by the number of publications published in 2017-2020. Learn more >

CiteScore 2020

$$0.5 = \frac{25{,}463 \text{ Citations } 2017 - 2020}{49{,}883 \text{ Documents } 2017 - 2020}$$

Calculated on 05 May, 2021

CiteScoreTracker 2021

$$0.5 = \frac{37{,}014 \text{ Citations to date}}{67{,}488 \text{ Documents to date}}$$

Last updated on 04 November, 2021 • Updated monthly

CiteScore rank 2020

Category	Rank	Percentile
Environmental Science General Environmental Science	#183/220	17th
Earth and Planetary Sciences General Earth and Planetary Sciences	#157/186	15th

View CiteScore methodology > CiteScore FAQ > Add CiteScore to your site

IOP Conference Series: Earth and Environmental Science

Table of contents

Volume 293
2019

‹ Previous issue Next issue ›

The 2nd International Conference on Natural Resources and Life Sciences (NRLS-2018) 23–24 August 2018, Ibis Styles Hotel, Surabaya, Indonesia

Accepted papers received: 17 May 2019
Published online: 01 July 2019

Open all abstracts

Preface

Papers

OPEN ACCESS 012004

Review on biogas from palm oil mill effluent (POME): Challenges and opportunities in Indonesia

A Rajani, Kusnadi, A Santosa, A Saepudin, S Gobikrishnan and D Andriani

+ Open abstract View article PDF

OPEN ACCESS 012005

Early detection of somaclonal variation in oil palm callus culture through cytological and SDS-PAGE protein analysis

A Sahara, Refilini, C Utomo and T Liwang

+ Open abstract View article PDF

OPEN ACCESS 012006

Genetic diversity of Indonesian soybean (*Glycine max* L. Merrill) germplasm based on morphological and microsatellite markers

A Sulistyo, F C Indriani, M J Mejaya, A N Sugiharto and J Agranoff

+ Open abstract View article PDF

OPEN ACCESS 012007

Fermentation quality of *Pennisetum purpureum* cv. Mott ensiled with *Lactobacillus plantarum* and sugarcane molasses in tropic

A Wahyudi, L Hendraningsih, Sutawi, R H Setyobudi and M Mel

+ Open abstract View article PDF

OPEN ACCESS 012008

The evaluation of estimated breeding value and the most probable producing ability for the basis selection of Ettawa crossbred goat (*Capra hircus* sp.) at Malang, East Java, Indonesia

A Winaya, Suyatno, P Coy and N Fauzi

+ Open abstract View article PDF

OPEN ACCESS 012009

Menthol from the stem and leaf *in-vitro Mentha piperita* Linn.

B K Wijaya, P H Hardjo and S Emantoko

+ Open abstract View article PDF

OPEN ACCESS 012010

A review: Biomass-based fuel pellet usage in biomass gasifier-an optimization on non-wood material

D Andriani, T D Atmaja, M Arifin, A Rajani and Kusnadi

+ Open abstract View article PDF

OPEN ACCESS 012011

Effectiveness of the use of organic waste as fertilizer and physical scarification of seeds on growth of seeds nila plants (*Indigofera* sp.)

D Roeswitawati, M Huda, D Indratmi and M Mel

+ Open abstract View article PDF

OPEN ACCESS 012012

Physiology and genotyping of adaptive and sensitive oil palm progenies under unwatered stress condition

D Yono, E Purwanti, A Sahara, Y A Nugroho, Z A Tanjung, R Aditama, C Dewi, A E Silhotang, C Utomo and T Liwang

+ Open abstract View article PDF

OPEN ACCESS 012013

Effect of phosphoric acid pretreatment on characterization of gelatin from broiler chicken (*Gallus gallus* domesticus L.) bones

D Yuliani, A Maunatin, A Jannah and H H Fauziyyah

+ Open abstract View article PDF

OPEN ACCESS 012015

Effectiveness the source of nitrogen from NO_3 and NH_4 for *Panicum maximum* Jacq. growth in saline soil

E D Purbajanti, P G Adinurani, T Turkadze, Z Vinčeković-Gašić and R H Setyobudi

OPEN ACCESS 012016

Anticancer potential from ethanol extract of *Zanthoxylum acanthopodium* DC. seed to against MCF-7 cell line

E V Arsita, D E Saragih and K Aldrin

OPEN ACCESS 012017

Long-term Proton Pump Inhibitors induces recurrent Urinary Tract Infections: A case study

F Colero and S T Anang

OPEN ACCESS 012018

Molecular docking, drug-likeness, and ADMET study of 1-benzyl-3-benzoylurea and its analogs against VEGFR-2

F Suhud, D H Tjahjono, T A Yunianta, G S Putra and J Setiawan

OPEN ACCESS 012019

Microcontroller application for pH and temperature control system in liquid sugar liquefaction process made from cassava *(Manihot esculanta* Crantz.)

H Santosa, Yullati, I J Mulyana, D S Sirait and S D Novitasari

OPEN ACCESS 012020

The effect of fermentation process on physical and chemical characteristics of pitaya *(Hylocereus polyrhiuzus* [F.A.C. Weber] Britton & Rose) stem flour

H Soedjatmiko, R Chrismasari and P H Hardjo

OPEN ACCESS 012021

The performance of three local rice (*Oryza sativa* L.) cultivar from East Kalimantan - Indonesia under drought stress at early seedling stage

I B M Artadana, I T Dewi and J Sukweenadhi

OPEN ACCESS 012022

Coffee reduced the production of neutrophil superoxide radical *in vitro*

I D A Susilawati, A Safaatin and J Burlakovs

OPEN ACCESS 012023

The potential of extract of *Zingiber zerumbet* (L.) Smith as a feed additive to improve the production performances and meat nutritional composition of broiler chickens

I D Rahayu, W Widodo, I Prihartini, A Winaya, L Zalizar, T Untari and M Mel

OPEN ACCESS 012024

Callus induction and *in vitro* mass culture of adventitious roots from leaf segment explants of *Dendropanax morbifera* Lev.

J Sukweenadhi, J Y Choi, Y J Kim, L Kalinaj, S Abid, J C Ahn and D C Yang

OPEN ACCESS 012029

The drought stress tolerance of physic nut (*Jatropha curcas* Linn.) genotypes

Maftuchah, I Z Fahmi, A Zainudin, A Ikhwan, Djumali and L K Kuan

+ Open abstract View article PDF

OPEN ACCESS 012030

Characterization of thermostable chitinase from *Bacillus licheniformis* B2

N Jayanthi, M G M Purwanto, R Chrisnasari, T Pantjajani, A Wahjudi and M Sugiarto

+ Open abstract View article PDF

OPEN ACCESS 012031

Antibacterial activity roll on deodorant with *Pluchea indica* (L.) leaf extract against *Staphylococcus epidermidis* (Evans 1916) *in vitro*

O Komala, I Y Wiendarlina and N Rizqiyana

+ Open abstract View article PDF

OPEN ACCESS 012032

Production potensial of sweet corn (*Zea mays* Linn. var. Saccharata Sturt) 'Bonanza' to different planting pattern and phosphorus sources

P G Adinurani, S Rahayu, L S Budi, S Pambudi and P Soni

+ Open abstract View article PDF

OPEN ACCESS 012033

Genome-wide identification of oil palm *(Elaeis guineensis* Jacq.) chitinases and their response to *Ganoderma boninense* Pat. infection

R Aditama, R Tryono, Z A Tanjung, C Utomo and T Liwang

+ Open abstract View article PDF

OPEN ACCESS 012034

Effect of new NPK fertilizer on lowland rice (*Oryza sativa* L.) growth

R Budiono, P G Adinurani and P Soni

+ Open abstract View article PDF

OPEN ACCESS 012035

Prospect of Fe non-heme on coffee flour made from solid coffee waste: Mini review

R H Setyobudi, L Zalizar, S K Wahono, W Widodo, A Wahyudi, M Mel, B Prabowo, Y Jani, Y A Nugroho, T Liwang *et al*

+ Open abstract View article PDF

OPEN ACCESS 012036

Biomass to methanol plant based on gasification of palm empty fruit bunch

R Heryadi, A S Uyun, E Yandri, S M Nur, K Abdullah and O Anne

+ Open abstract View article PDF

OPEN ACCESS 012037

Review article: Myopia - Genetically inherited or environmental influences

S E E Tjoa and S E D Putra

+ Open abstract View article PDF