Fabrication of Cellulose Acetate Film From Oil Palm Empty Fruit Bunch (OP-EFB) and Cytotoxicity Evaluation

Dasmawati Mohamad1*, Wan Suzaini Wan Hamzah1, Wan Rosli Wan Daud2, Zainul Ahmad Rajion1, Wan Zaripah Wan Bakar1 and Mazlan Ibrahim2

1School of Dental Sciences, 2School of Industrial Technology, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia
*E-mail: dasmawati@kck.usm.my

ABSTRACT
The aims of this study were to fabricate cellulose acetate (CA) film from oil palm empty fruit bunch (OP-EPB), as well as to characterize and evaluate their biocompatibility. Several processes were carried out, and these included prehydrolysis-soda method, chlorine free bleaching method, including oxygen, ozone and peroxide, to produce the cellulose pulp. Then, a liquid phase acetylation method was applied through acetic acid-acetic anhydride-sulphuric acid. Triethyl citrate (TEC) ester was used as additive at different percentages of 10, 20, 30 and 40 wt%. The film produced was characterized by FTIR to identify the functional group of the CA film and their tensile properties were further characterized. Biocompatibility of the film was evaluated using cytotoxicity test. Stem cell derived from human deciduous teeth (SHED) was used with MTS assay. The results showed at 30% of TEC, the tensile strength and elongation of CA (OP-EFB) film was at the optimum and is therefore suitable to be used in dental application. The cytotoxicity evaluated showed that the fabricated CA (OP-EFB) films were non-toxic up to the concentration tested, and are thus compatible with SHED.

Keywords: Oil palm empty fruit bunch, cellulose acetate film, cytotoxicity

INTRODUCTION
In Malaysia, enormous volumes of agricultural wastes, empty fruit bunch (EFB) containing cellulosic fibres are generated annually (Suhaimi & Ong, 2001). Many of these wastes are allowed to rot away unutilized. These agricultural wastes can actually be maximized their utilization, as the focus of this study is, in the production of pulp for papermaking and conversion to cellulose derivatives, specifically cellulose acetate (CA). The application of CA is widely used in photography film, automotive coatings, selective filtration membranes in medicine, and also in dental field. The advantages of CA film, such as being tough, with good dimensional stability and optical properties, made it suitable to be used in dental field. Due to its flexible properties, CA film is usually used to assist dentists while performing tooth restoration. In the fabrication of CA, which is a thermoplastic material (also referred to as bioplastic), must be modified to make it suitable for matrix polymers for commercial composite application. Plastisizers are widely used in the plastic industry to improve their processibility, flexibility and ductility properties. Conventionally, cellulose ester plastics are plasticized with a petroleum–derived phthalate plasticizer, which is not environmental friendly. Mohanty et al. carried out a study on cellulose acetate plasticized with varying concentrations

Received: 18 April 2011
Accepted: 12 September 2011
*Corresponding Author
of an eco-friendly triethyl citrate (TEC) plasticizer (2003). The results were very promising and plasticized cellulose acetate was found to be processable at 170-180°C, i.e. approximately 50°C below the melting point of neat cellulose acetate.

The aims of this study were to fabricate cellulose acetate (CA) film from oil palm empty fruit bunches (OP-EPB) and investigate the effects of TEC plasticizer on the tensile properties of the resulting CA (OP-EPB) film. In addition, cytotoxicity was also evaluated. TEC was used as the plasticizer at different compositions. The CA films were characterized by Fourier Transform Infra-Red Spectroscopy (FTIR). Both the tensile strength and percentage elongation were also evaluated. Since the fabricated CA film is intended to be used in dental application and applied for oral purposes, the biocompatibility assessment is needed. One of the criteria for biocompatibility is that the material is not toxic to cells. Therefore, the fabricated CA films underwent a cytotoxicity test using MTS assay and the stem cell from human deciduous teeth (SHED) was also used. Vital staining was carried out as proposed by the National Guidelines for cytotoxicity test ISO 10993.

**MATERIALS AND METHODS**

*Materials*

Oil palm empty fruit bunches (OP-EFB) were collected from SABUTEK (M) Sdn. Bhd. in form of fibrous strands and used as raw material. TEC plasticizer was purchased from Merck Company.

*Preparation of Cellulose Acetate Film*

*Preparation of the raw material*

The raw material (OP-EFB) was cut into pieces and boiled with distilled water for 60 minutes at 170°C, before the pulp underwent soda pulping using sodium hydroxide solution for 100 min at 160°C to remove non-cellulosic materials. Later, it was washed with water and air dried.

*Bleaching*

The chlorine-free bleaching process studied here includes three sequential steps of oxygen, ozone and hydrogen peroxide bleaching. Oxygen (O₂) bleaching was carried out under alkaline conditions by addition of 1% NaOH (w/w) aqueous solution. Magnesium sulphate (0.1%) was also added as a protection reagent for cellulose. For ozone (O₃) bleaching, the ozone was produced from the supplied oxygen gas and was mixed with pulp. Total reaction time was 2.5 min with occasional mixing at 60 rpm. For hydrogen peroxide (H₂O₂) bleaching, the pulp was placed in a plastic bag and NaOH MgSO₄ was added according to the weight of the pulp (Tanaka et al., 2002). The reaction was then carried out at 60 °C for 60 min in a water bath.

*Acetylation of Cellulose*

The cellulose acetate is usually produced by treating cellulose with acetic acid first as activation phase. To 5 parts of pulp, 90 ml of acetic acid, and 0.5 ml of sulphuric acid were added and stirred vigorously. After one min, 25 ml of acetic anhydride was added and the stirring was continued. Later, an equal volume of water was added into the reaction mixture to precipitate CA. The degree of acetylation of cellulose was determined using a standard method based on ASTM D-871-61T. The degree of acetylation found was 37.41%, and thus, the degree of substitution of CA was 2.2.
Preparation of Films
CA (OP-EFB) film was made using a casting method. First, a mixture solution of CA in acetone at 16 % (w/v) was prepared. Plasticizer material, TEC, was then dissolved in the solvent mixture at different compositions (namely CA1, CA2, CA3, and CA4) as in Table 1 and stirred for 48 h. Next, the mixtures were centrifuged at 3000 rpm for 1 h. The CA solution of 12 ml was slowly and evenly poured into a petry dish of 8 cm in diameter and was dried in a dessicator for 24 h. The thickness of the CA film obtained was approximately 0.1 mm.

TABLE 1
CA at different compositions of plasticizer

<table>
<thead>
<tr>
<th>Specimen</th>
<th>CA/Acetone solution (ml) (w/v%)</th>
<th>Plasticizer (ml) Composition of TEC (wt%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA1</td>
<td>16</td>
<td>10</td>
</tr>
<tr>
<td>CA2</td>
<td>16</td>
<td>20</td>
</tr>
<tr>
<td>CA3</td>
<td>16</td>
<td>30</td>
</tr>
<tr>
<td>CA4</td>
<td>16</td>
<td>40</td>
</tr>
</tbody>
</table>

Characterization

FTIR Spectroscopy
The IR spectra of various specimens of the CA films were taken using Nicolet Impact 400 Fourier Transform Infrared spectrophotometer using KBr pellet. For each CA at different compositions of plasticizer, three specimens (n=3) were used for the FTIR evaluation. The peaks of C=O, C=C and C-O were taken and compared with a standard CA powder and also experimental CA (OP-EFB) powders.

Tensile Strength and Elongation
The tests were carried out using a Hounsfield TX0201 Tensile Testing System (H10KS model) according to the ASTM D 882. The thickness of the specimens for the tensile evaluation was maintained at approximately 0.1 mm and the length between the grips was set at 30mm. The measurements of CA films were made at three specimens, (n=3) for each CA at different compositions of plasticizer.

Cytotoxicity Evaluation
The national guidelines for cytotoxicity test ISO /EN 10993-5 are followed for the biocompatibility evaluation. The MTS assay of [3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt] was used for the cytotoxicity evaluation with the CA(OP-EFB) extracts on the stem cell derived from human decidua teeth (SHED). This cytotoxicity test was carried out according to the ISO 10993-5 standard (ASTM Standard D882). The SHED were initially cultured at cell density 5 x 10^3 cells/cm for 24 hours at 37 °C in 96 well plates. The medium was replaced with the sample extract. However, only one representative sample was used for this cytotoxicity evaluation. The plates were incubated in a CO_2 incubator for 72 hours. Next, 5mg MTS powder was measured and mixed thoroughly with 1 ml dulbecco’s phosphate buffered saline.
(DPBS) in a small sterile universal tube. After being thoroughly mixed, the MTS solution was filtered. 10µl of the MTS solution was added into all the 96 wells after 72 hours of incubation. The plates were further incubated for 2-4 hours in 5% CO₂ incubator. After that, the culture medium and excessive MTS solution were removed by inversion and blotted carefully on tissue paper. Next, 100µl of dimethylsulfoxide (DMSO) was added into each well and gently shaken for 5 minutes to achieve complete dissolution. Finally, an Elisa reader (TECAN) was used to read the absorbance at the reference and test wavelengths of 600nm and 570nm. The cell viability percentages were calculated according to the following equation:

\[
\text{% Cell viability} = \frac{OD_{\text{sample}}}{OD_{\text{control}}} \times 100\%
\]

where, OD= Optical density.

RESULTS AND DISCUSSION

The degree of the substitution of the CA found was 2.2. In general, those cellulose acetates with acetyl substitution numbers of 2.2 or less are biodegradable in soil and marine environment and are therefore suitable for composting compared with higher substitution numbers from 2.2 to 3.0. Hence, the CA (OP-EFB) film produced was confirmed as biodegradable.

The CA (OP-EFB) films of all the different percentages of TEC plastisizers were confirmed using the FTIR evaluations (Table 2). The present of the functional group C=O, C=C, and C-O indicates that the structure of CA was confirmed, as compared to the commercial CA which was done only on the powder form (Table 3). The acetate groups were found in each sample; however, the peak value was different from the control group since they were in the powder form. Nonetheless, there was not much difference between the peaks in the powder form of the commercial CA and (OP-EFB) CA, as shown in Fig. 1.
TABLE 2  
The FTIR peak results of the CA (OP-EFB) films

<table>
<thead>
<tr>
<th>Specimen</th>
<th>C=O (cm$^{-1}$)</th>
<th>C=C (cm$^{-1}$)</th>
<th>C-O (acetate) (cm$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA1</td>
<td>1711.51 (0.85)</td>
<td>1637.41 (0.20)</td>
<td>1071.73 (15.45)</td>
</tr>
<tr>
<td>CA2</td>
<td>1736.75 (22.75)</td>
<td>1636.71 (1.26)</td>
<td>1071.69 (2.17)</td>
</tr>
<tr>
<td>CA3</td>
<td>1765.42 (1.13)</td>
<td>1636.15 (0.33)</td>
<td>1087.22 (34.00)</td>
</tr>
<tr>
<td>CA4</td>
<td>1753.85 (16.10)</td>
<td>1636.27 (0.13)</td>
<td>1152.63 (94.75)</td>
</tr>
</tbody>
</table>

*Note: The reported are mean values with their standard deviation in brackets.*

The FTIR analysis identified all the important peaks present in the sample CA (OP-EFB), which were nearly similar to the peaks that appeared in the CA commercial. A summary of the peak wave number for all the functional groups C=O, C=C, and C-O present in CA commercial and CA (OP-EFB) is tabulated in Table 3.

TABLE 3  
The FTIR peak results of CA in powder form

<table>
<thead>
<tr>
<th>Specimen</th>
<th>C=O (cm$^{-1}$)</th>
<th>C=C (cm$^{-1}$)</th>
<th>C-O (acetate) (cm$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA (Standard commercial)</td>
<td>1754.56</td>
<td>1638.57</td>
<td>1239.53</td>
</tr>
<tr>
<td>CA (OP-EFB)</td>
<td>1754.51</td>
<td>1643.01</td>
<td>1243.31</td>
</tr>
</tbody>
</table>

In the tensile properties evaluation, the results presented in Table 4 show an increasing trend of the tensile strength of CA (OP-EFB) films with a decreasing composition of TEC plasticizer. Although 10% of TEC has the highest value of tensile strength, the CA1 film produced was brittle, not very flexible, and it tended to wrinkle. This is shown by having the lowest elongation of 6.51%. At 20% of TEC, CA2 film also has the same features as those of CA1, but with improved elongation properties. On the other hand, CA3 gave a tensile strength of about 15 MPa but the film was very flexible and it showed the highest elongation. An optimum balance between the tensile strength and elongation is depending on the CA application. As the fabricated CA was intended to be used in dental application, which is supposed to bend easily around a tooth, the CA3 composition is therefore suitable. For CA4 film, the tensile strength was the lowest as compared to the other composition of plasticizer. The result found is in agreement with the finding by Mohanty *et al.* (2003).

TABLE 4  
The effect of different compositions of TEC on the tensile strength and elongation of CA (OP-EFB) films

<table>
<thead>
<tr>
<th>Specimen</th>
<th>CA/Acetone (w/v%)</th>
<th>Composition of TEC (wt%)</th>
<th>Tensile strength (MPa)</th>
<th>Elongation (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA1</td>
<td>16</td>
<td>10</td>
<td>27.04 (1.98)</td>
<td>6.51</td>
</tr>
<tr>
<td>CA2</td>
<td>16</td>
<td>20</td>
<td>21.11 (0.22)</td>
<td>10.67</td>
</tr>
<tr>
<td>CA3</td>
<td>16</td>
<td>30</td>
<td>15.71 (1.13)</td>
<td>10.76</td>
</tr>
<tr>
<td>CA4</td>
<td>16</td>
<td>40</td>
<td>7.48 (0.40)</td>
<td>10.61</td>
</tr>
</tbody>
</table>

*Note: Reported are mean values with their standard deviation in brackets.*
However, to achieve the tensile strength of CA (OP-EFB) that is similar to the commercial CA films, more extensive research is needed. Their tensile strength is about 100–140 MPa (Zugenmaier & Peter, 2004). Many factors may contribute to their high tensile strength, such as different origins of cellulose, molecular weight, purity, and different methods of film preparation.

Fig. 2: The effect of TEC plasticizer on the tensile strength and elongation of the CA films

In this study, CA (OP-EFB) was found to be not fully dissolved in acetone, which resulted in the obtained supernatant after centrifuged had thinner film compared to the commercial CA. Hence, different types of solvent need to be further explored. TEC composition is also important in the final product of CA film. The final application of the CA film is dependent upon the plasticizer composition. Other factors, such as drying method, have also been reported to affect the quality of the film. Meanwhile, the drying process plays an important role in affecting the wrinkles of the fabricated film.

Fig. 3 presents the cell viability of the CA films in function of extract concentration. IC_{50} (50% Inhibitory Concentration) endpoint was used to evaluate the cytotoxicity effects of the materials at different concentrations applied. From the figure, the percentages of cell viability decrease when the CA extraction concentration is high. However, at the concentrations of 150mg/ml onwards, the curve line becomes a plateau at slightly above 50%. Therefore, the fabricated CA can be considered as non-toxic. However, further test on biocompatibility is still needed, such as genotoxicity and Ames test.

Fig. 3: The percentage of the cell viability of CA film extraction on SHED
CONCLUSIONS

In this study, the CA produced from OP-EFB obtained a DS of 2.2. Therefore, it is possible to use TEC as it is eco-friendly and promoting green technology. However, increasing the amount of TEC plasticizer significantly reduced the tensile strength. The optimum balance of strength and stiffness of the CA film at 30 (wt%) plasticizer is found to be suitable for dental application. The cytotoxicity evaluation provides evidence which indicates that CA (OP-EFB) is non-toxic to the SHED, up to the concentration tested.

ACKNOWLEDGEMENT

The authors would like to thank all the technicians from Craniofacial Science Laboratory, School of Dental Sciences, Universiti Sains Malaysia who were involved in this project. This study was supported by Yayasan FELDA with the following grant (304/PPSG/6150087/Y104).

REFERENCES


The Editorial Board of the Journal of Science and Technology wishes to thank the following for acting as referees for manuscripts published in this issue of JST.

Abu Hassan Shaari Md Nor
Agus Arsad
Ahmad Shukri Muhammad Noor
Aida Isma Mohd Idris
Amru Nasrulhaq Boyce
Ashutosh Kumar Singh
Azmi Abdul Wahab
Badronnisa Yusof
Chantara Thevy Ratnam
Cheong Ai Theng
Desa Ahmad
Farah Saleena Taip
Hang Tuah Baharudin, BT
Intan Salwani Ahamad
M Iqbal Saripan
Mansor Hashim
Mariana Nor Shamsudin
Mohammad Esmail Arasteh Rad
Miralini Kandiah
Mohd Shamsul Bin Anuar
Rahmita Wirza OK Rahmat
Razali Yaakob
Rohaya Latip
Rosnita A Talib
Rozita Omar
Salmiaton Ali
Shafreeza Sobri
Sharipah Soaad Syed Yahaya
Sidek Ab Aziz
Siti Khairunniza Bejo
Siti Mazlina Mustapa Kamal
Siti Suri Arshad
Somayeh Habibi
Teng Beng Ti
Wan Ishak Wan Ismail
Wan Mahmood Mat Yunus
Zurina Zainal Abidin

Special Acknowledgement
The JST Editorial Board gratefully acknowledges the assistance of Doreen Dillah, who served as the English language editor for this issue.

While every effort has been made to include a complete list of referees for the period stated above, however if any name(s) have been omitted unintentionally or spelt incorrectly, please notify the Executive Editor, Pertanika Journals at ndeeps@admin.upm.edu.my.

Any inclusion or exclusion of name(s) on this page does not commit the Pertanika Editorial Office, nor the UPM Press or the University to provide any liability for whatsoever reason.
Pertanika

Our goal is to bring high quality research to the widest possible audience

Journal of Science & Technology

INSTRUCTIONS TO AUTHORS
(Manuscript Preparation & Submission Guidelines)
Revised January 2012

We aim for excellence, sustained by a responsible and professional approach to journal publishing. We value and support our authors in the research community.

Please read the guidelines and follow these instructions carefully; doing so will ensure that the publication of your manuscript is as rapid and efficient as possible. The Editorial Board reserves the right to return manuscripts that are not prepared in accordance with these guidelines.

About the Journal
Pertanika is an international peer-reviewed journal devoted to the publication of original papers, and it serves as a forum for practical approaches to improving quality in issues pertaining to tropical agriculture and its related fields. Pertanika began publication in 1978 as Journal of Tropical Agricultural Science. In 1992, a decision was made to streamline Pertanika into three journals to meet the need for specialised journals in areas of study aligned with the interdisciplinary strengths of the university. The revamped Journal of Science and Technology (JST) is now focusing on research in science and engineering, and its related fields. Other Pertanika series include Journal of Tropical Agricultural Science (JTAS); and Journal of Social Sciences and Humanities (JSSH).

JST is published in English and it is open to authors around the world regardless of the nationality. It is currently published two times a year i.e. in January and July.

Goal of Pertanika
Our goal is to bring the highest quality research to the widest possible audience.

Quality
We aim for excellence, sustained by a responsible and professional approach to journal publishing. Submissions are guaranteed to receive a decision within 12 weeks. The elapsed time from submission to publication for the articles averages 5-6 months.

Indexing of Pertanika
Pertanika is now over 33 years old; this accumulated knowledge has resulted in Pertanika JST being indexed in SCOPUS (Elsevier), EBSCO and DOAJ.

Future vision
We are continuously improving access to our journal archives, content, and research services. We have the drive to realise exciting new horizons that will benefit not only the academic community, but society itself.

We also have views on the future of our journals. The emergence of the online medium as the predominant vehicle for the ‘consumption’ and distribution of much academic research will be the ultimate instrument in the dissemination of the research news to our scientists and readers.

Aims and scope
Pertanika Journal of Science and Technology aims to provide a forum for high quality research related to science and engineering research. Areas relevant to the scope of the journal include: bioinformatics, bioscience, biotechnology and bio-molecular sciences, chemistry, computer science, ecology, engineering, engineering design, environmental control and management, mathematics and statistics, medicine and health sciences, nanotechnology, physics, safety and emergency management, and related fields of study.
Editorial Statement

*Pertanika* is the official journal of Universiti Putra Malaysia. The abbreviation for *Pertanika* Journal of Science & Technology is *Pertanika J. Sci. Technol.*

Guidelines for Authors

Publication policies

*Pertanika* policy prohibits an author from submitting the same manuscript for concurrent consideration by two or more publications. It prohibits as well publication of any manuscript that has already been published either in whole or substantial part elsewhere. It also does not permit publication of manuscript that has been published in full in Proceedings. Please refer to *Pertanika*’s Code of Ethics for full details.

Editorial process

Authors are notified on receipt of a manuscript and upon the editorial decision regarding publication.

**Manuscript review:** Manuscripts deemed suitable for publication are sent to the Editorial Board members and/or other reviewers. We encourage authors to suggest the names of possible reviewers. Notification of the editorial decision is usually provided within eight to ten weeks from the receipt of manuscript. Publication of solicited manuscripts is not guaranteed. In most cases, manuscripts are accepted conditionally, pending an author’s revision of the material.

**Author approval:** Authors are responsible for all statements in articles, including changes made by editors. The liaison author must be available for consultation with an editor of *The Journal* to answer questions during the editorial process and to approve the edited copy. Authors receive edited typescript (not galley proofs) for final approval. Changes **cannot** be made to the copy after the edited version has been approved.

Please direct all inquiries, manuscripts, and related correspondence to:

The Executive Editor  
*Pertanika* Journals, UPM Press  
Office of the Deputy Vice Chancellor (R&I)  
IDEA Tower II, UPM-MTDC Technology Centre  
Universiti Putra Malaysia  
43400 UPM, Serdang, Selangor  
Malaysia  
Phone: + (603) 8947 1622  
ndeepps@admin.upm.edu.my

or visit our website at [http://www.pertanika.upm.edu.my/](http://www.pertanika.upm.edu.my/) for further information.

Manuscript preparation

*Pertanika* accepts submission of mainly four types of manuscripts. Each manuscript is classified as **regular** or **original** articles, **short communications**, **reviews**, and proposals for **special issues**. Articles must be in **English** and they must be competently written and argued in clear and concise grammatical English. Acceptable English usage and syntax are expected. Do not use slang, jargon, or obscure abbreviations or phrasing. Metric measurement is preferred; equivalent English measurement may be included in parentheses. Always provide the complete form of an acronym/abbreviation the first time it is presented in the text. Contributors are strongly recommended to have the manuscript checked by a colleague with ample experience in writing English manuscripts or an English language editor.

Linguistically hopeless manuscripts will be rejected straightaway (e.g., when the language is so poor that one cannot be sure of what the authors really mean). This process, taken by authors before submission, will greatly facilitate reviewing, and thus publication if the content is acceptable.

The instructions for authors must be followed. Manuscripts not adhering to the instructions will be returned for revision without review. Authors should prepare manuscripts according to the guidelines of *Pertanika*.

1. **Regular article**  
**Definition:** Full-length original empirical investigations, consisting of introduction, materials and methods, results and discussion, conclusions. Original work must provide references and an explanation on research findings that contain new and significant findings.

**Size:** Should not exceed 5000 words or 8-10 printed pages (excluding the abstract, references, tables and/or figures). One printed page is roughly equivalent to 3 type-written pages.
2. Short communications

Definition: Significant new information to readers of the Journal in a short but complete form. It is suitable for the publication of technical advance, bioinformatics or insightful findings of plant and animal development and function.

Size: Should not exceed 2000 words or 4 printed pages, is intended for rapid publication. They are not intended for publishing preliminary results or to be a reduced version of Regular Papers or Rapid Papers.

3. Review article

Definition: Critical evaluation of materials about current research that had already been published by organizing, integrating, and evaluating previously published materials. Re-analyses as meta-analysis and systemic reviews are encouraged. Review articles should aim to provide systemic overviews, evaluations and interpretations of research in a given field.

Size: Should not exceed 4000 words or 7-8 printed pages.

4. Special issues

Definition: Usually papers from research presented at a conference, seminar, congress or a symposium.

Size: Should not exceed 5000 words or 8-10 printed pages.

5. Others

Definition: Brief reports, case studies, comments, Letters to the Editor, and replies on previously published articles may be considered.

Size: Should not exceed 2000 words or up to 4 printed pages.

With few exceptions, original manuscripts should not exceed the recommended length of 6 printed pages (about 18 typed pages, double-spaced and in 12-point font, tables and figures included). Printing is expensive, and, for the Journal, postage doubles when an issue exceeds 80 pages. You can understand then that there is little room for flexibility.

Long articles reduce the Journal's possibility to accept other high-quality contributions because of its 80-page restriction. We would like to publish as many good studies as possible, not only a few lengthy ones. (And, who reads overly long articles anyway?) Therefore, in our competition, short and concise manuscripts have a definite advantage.

Format

The paper should be formatted in one column format with at least 4cm margins and double spacing throughout. Authors are advised to use Times New Roman 12-point font. Be especially careful when you are inserting special characters, as those inserted in different fonts may be replaced by different characters when converted to PDF files. It is well known that ‘µ’ will be replaced by other characters when fonts such as ‘Symbol’ or ‘Mincho’ are used.

A maximum of eight keywords should be indicated below the abstract to describe the contents of the manuscript. Leave a blank line between each paragraph and between each entry in the list of bibliographic references. Tables should preferably be placed in the same electronic file as the text. Authors should consult a recent issue of the Journal for table layout.

Every page of the manuscript, including the title page, references, tables, etc. should be numbered. However, no reference should be made to page numbers in the text; if necessary, one may refer to sections. Underline words that should be in italics, and do not underline any other words.

We recommend that authors prepare the text as a Microsoft Word file.

1. Manuscripts in general should be organised in the following order:

   ○ Page 1: Running title. (Not to exceed 60 characters, counting letters and spaces). This page should only contain the running title of your paper. The running title is an abbreviated title used as the running head on every page of the manuscript.

     In addition, the Subject areas most relevant to the study must be indicated on this page. Select one or two subject areas (refer to the Scope Form).

     A list of number of black and white / colour figures and tables should also be indicated on this page. Figures submitted in color will be printed in colour. See “5. Figures & Photographs” for details.

   ○ Page 2: Author(s) and Corresponding author information. This page should contain the full title of your paper with name(s) of all the authors, institutions and corresponding author’s name, institution and full address (Street address, telephone number (including extension), hand phone number, fax number and e-mail address) for editorial correspondence. The names of the authors must be abbreviated following the international naming convention. e.g. Salleh, A.B., Tan, S.G., or Sapuan, S.M.
Authors' addresses. Multiple authors with different addresses must indicate their respective addresses separately by superscript numbers:

George Swan1 and Nayan Kanwal2
1Department of Biology, Faculty of Science, Duke University, Durham, North Carolina, USA. 
2Research Management Centre, Universiti Putra Malaysia, Serdang, Malaysia.

- Page 3: This page should repeat the full title of your paper with only the Abstract (the abstract should be less than 250 words for a Regular Paper and up to 100 words for a Short Communication). Keywords must also be provided on this page (Not more than eight keywords in alphabetical order).

- Page 4 and subsequent pages: This page should begin with the Introduction of your article and the rest of your paper should follow from page 5 onwards.

Abbreviations. Define alphabetically, other than abbreviations that can be used without definition. Words or phrases that are abbreviated in the introduction and following text should be written out in full the first time that they appear in the text, with each abbreviated form in parenthesis. Include the common name or scientific name, or both, of animal and plant materials.

Footnotes. Current addresses of authors if different from heading.

2. Text. Regular Papers should be prepared with the headings Introduction, Materials and Methods, Results and Discussion, Conclusions in this order. Short Communications should be prepared according to "8. Short Communications," below.

3. Tables. All tables should be prepared in a form consistent with recent issues of Pertanika and should be numbered consecutively with Arabic numerals. Explanatory material should be given in the table legends and footnotes. Each table should be prepared on a separate page. (Note that when a manuscript is accepted for publication, tables must be submitted as data - .doc, .rtf, Excel or PowerPoint file- because tables submitted as image data cannot be edited for publication.)

4. Equations and Formulae. These must be set up clearly and should be typed triple spaced. Numbers identifying equations should be in square brackets and placed on the right margin of the text.

5. Figures & Photographs. Submit an original figure or photograph. Line drawings must be clear, with high black and white contrast. Each figure or photograph should be prepared on a separate sheet and numbered consecutively with Arabic numerals. Appropriate sized numbers, letters and symbols should be used, no smaller than 2 mm in size after reduction to single column width (85 mm), 1.5-column width (120 mm) or full 2-column width (175 mm).

Failure to comply with these specifications will require new figures and delay in publication. For electronic figures, create your figures using applications that are capable of preparing high resolution TIFF files acceptable for publication. In general, we require 300 dpi or higher resolution for coloured and half-tone artwork and 1200 dpi or higher for line drawings. For review, you may attach low-resolution figures, which are still clear enough for reviewing, to keep the file of the manuscript under 5 MB. Illustrations may be produced at extra cost in colour at the discretion of the Publisher; the author could be charged Malaysian Ringgit 50 for each colour page.

6. References. Literature citations in the text should be made by name(s) of author(s) and year. For references with more than two authors, the name of the first author followed by ‘et al.’ should be used.

Swan and Kanwal (2007) reported that …
The results have been interpreted (Kanwal et al. 2009).

- References should be listed in alphabetical order, by the authors’ last names. For the same author, or for the same set of authors, references should be arranged chronologically. If there is more than one publication in the same year for the same author(s), the letters ‘a’, ‘b’, etc., should be added to the year.

- When the authors are more than 11, list 5 authors and then et al.

- Do not use indentations in typing References. Use one line of space to separate each reference. The name of the journal should be written in full. For example:


In case of citing an author(s) who has published more than one paper in the same year, the papers should be distinguished by addition of a small letter as shown above, e.g. Jalaludin (1997a); Jalaludin (1997b).

Unpublished data and personal communications should not be cited as literature citations, but given in the text in parentheses. ‘In press’ articles that have been accepted for publication may be cited in References. Include in the citation the journal in which the ‘in press’ article will appear and the publication date, if a date is available.

7. Examples of other reference citations:


8. Short Communications should include Introduction, Materials and Methods, Results and Discussion, Conclusions in this order. Headings should only be inserted for Materials and Methods. The abstract should be up to 100 words, as stated above. Short Communications must be 5 printed pages or less, including all references, figures and tables. References should be less than 30. A 5 page paper is usually approximately 3000 words plus four figures or tables (if each figure or table is less than 1/4 page).

*Authors should state the total number of words (including the Abstract) in the cover letter. Manuscripts that do not fulfill these criteria will be rejected as Short Communications without review.

**STYLE OF THE MANUSCRIPT**

Manuscripts should follow the style of the latest version of the Publication Manual of the American Psychological Association (APA). The journal uses British spelling and authors should therefore follow the latest edition of the Oxford Advanced Learner’s Dictionary.

**SUBMISSION OF MANUSCRIPTS**

All articles should be submitted electronically using the ScholarOne web-based system. ScholarOne, a Thomson Reuters product provides comprehensive workflow management systems for scholarly journals. For more information, go to our web page and click “Online Submission”.

Alternatively, you may send your electronic files (manuscript, along with the Form BR 25 comprising Declaration, Referral A and Scope form) together with a cover letter directly to the Executive Editor. If the files are too big to email, mail a CD of your files. The forms and the sample of the cover letter are available from the Pertanika’s home page at [http://www.pertanika.upm.edu.my/](http://www.pertanika.upm.edu.my/) or from the Executive Editor’s office upon request.

All articles submitted to the journal must comply with these instructions. Failure to do so will result in return of the manuscript and possible delay in publication.

Please do not submit manuscripts to the editor-in-chief or to UPM Press directly. All manuscripts must be submitted through the executive editor’s office to be properly acknowledged and rapidly processed:

Dr. Nayan KANWAL
The Executive Editor
Pertanika Journals, UPM Press
Office of the Deputy Vice Chancellor (R&I)
IDEA Tower II, UPM-MTDC Technology Centre
Universiti Putra Malaysia
43400 UPM, Serdang, Selangor, Malaysia
email: ndeeps@admin.upm.edu.my; tel: + 603-8947 1622

Cover letter

All submissions must be accompanied by a cover letter detailing what you are submitting. Papers are accepted for publication in the journal on the understanding that the article is original and the content has not been published or submitted for publication elsewhere. This must be stated in the cover letter.

The cover letter must also contain an acknowledgement that all authors have contributed significantly, and that all authors are in agreement with the content of the manuscript.
The cover letter of the paper should contain (i) the title; (ii) the full names of the authors; (iii) the addresses of the institutions at which the work was carried out together with (iv) the full postal and email address, plus facsimile and telephone numbers of the author to whom correspondence about the manuscript should be sent. The present address of any author, if different from that where the work was carried out, should be supplied in a footnote.

As articles are double-blind reviewed, material that might identify authorship of the paper should be placed on a cover sheet.

Peer review

In the peer-review process, three referees independently evaluate the scientific quality of the submitted manuscripts. The Journal uses a double-blind peer-review system. Authors are encouraged to indicate in referral form A the names of three potential reviewers, but the editors will make the final choice. The editors are not, however, bound by these suggestions.

Manuscripts should be written so that they are intelligible to the professional reader who is not a specialist in the particular field. They should be written in a clear, concise, direct style. Where contributions are judged as acceptable for publication on the basis of content, the Editor or the Publisher reserves the right to modify the typescripts to eliminate ambiguity and repetition and improve communication between author and reader. If extensive alterations are required, the manuscript will be returned to the author for revision.

The editorial review process

What happens to a manuscript once it is submitted to Pertanika? Typically, there are seven steps to the editorial review process:

1. The executive editor and the editorial board examine the paper to determine whether it is appropriate for the journal and should be reviewed. If not appropriate, the manuscript is rejected outright and the author is informed.

2. The executive editor sends the article-identifying information having been removed, to three reviewers. Typically, one of these is from the Journal’s editorial board. Others are specialists in the subject matter represented by the article. The executive editor asks them to complete the review in three weeks and encloses two forms: (a) referral form B and (b) reviewer’s comment form along with reviewer’s guidelines. Comments to authors are about the appropriateness and adequacy of the theoretical or conceptual framework, literature review, method, results and discussion, and conclusions. Reviewers often include suggestions for strengthening of the manuscript. Comments to the editor are in the nature of the significance of the work and its potential contribution to the literature.

3. The executive editor, in consultation with the editor-in-chief, examines the reviews and decides whether to reject the manuscript, invite the author(s) to revise and resubmit the manuscript, or seek additional reviews. Final acceptance or rejection rests with the Editorial Board, who reserves the right to refuse any material for publication. In rare instances, the manuscript is accepted with almost no revision. Almost without exception, reviewers’ comments (to the author) are forwarded to the author. If a revision is indicated, the editor provides guidelines for attending to the reviewers’ suggestions and perhaps additional advice about revising the manuscript.

4. The authors decide whether and how to address the reviewers’ comments and criticisms and the editor’s concerns. The authors submit a revised version of the paper to the executive editor along with specific information describing how they have answered the concerns of the reviewers and the editor.

5. The executive editor sends the revised paper out for review. Typically, at least one of the original reviewers will be asked to examine the article.

6. When the reviewers have completed their work, the executive editor in consultation with the editorial board and the editor-in-chief examine their comments and decide whether the paper is ready to be published, needs another round of revisions, or should be rejected.

7. If the decision is to accept, the paper is sent to that Press and the article should appear in print in approximately two to three months. The Publisher ensures that the paper adheres to the correct style (in-text citations, the reference list, and tables are typical areas of concern, clarity, and grammar). The authors are asked to respond to any queries by the Publisher. Following these corrections, page proofs are mailed to the corresponding authors for their final approval. At this point, only essential changes are accepted. Finally, the article appears in the pages of the Journal and is posted on-line.

English language editing

Authors are responsible for the linguistic accuracy of their manuscripts. Authors not fully conversant with the English language should seek advice from subject specialists with a sound knowledge of English. The cost will be borne by the author, and a copy of the certificate issued by the service should be attached to the cover letter.

Note When your manuscript is received at Pertanika, it is considered to be in its final form. Therefore, you need to check your manuscript carefully before submitting it to the executive editor.
Author material archive policy
Authors who require the return of any submitted material that is rejected for publication in the journal should indicate on the cover letter. If no indication is given, that author’s material should be returned, the Editorial Office will dispose of all hardcopy and electronic material.

Copyright
Authors publishing the Journal will be asked to sign a declaration form. In signing the form, it is assumed that authors have obtained permission to use any copyrighted or previously published material. All authors must read and agree to the conditions outlined in the form, and must sign the form or agree that the corresponding author can sign on their behalf. Articles cannot be published until a signed form has been received.

Lag time
The elapsed time from submission to publication for the articles averages 5-6 months. A decision of acceptance of a manuscript is reached in 2 to 3 months (average 9 weeks).

Back issues
Single issues from current and recent volumes are available at the current single issue price from UPM Press. Earlier issues may also be obtained from UPM Press at a special discounted price. Please contact UPM Press at penerbit@putra.upm.edu.my or you may write for further details at the following address:

UPM Press
Universiti Putra Malaysia
43400 UPM, Serdang
Selangor Darul Ehsan
Malaysia
Pertanika is an international peer-reviewed leading journal in Malaysia which began publication in 1978. The journal publishes in three different areas — Journal of Tropical Agricultural Science (JTAS); Journal of Science and Technology (JST); and Journal of Social Sciences and Humanities (JSSH).

JTAS is devoted to the publication of original papers that serve as a forum for practical approaches to improving quality in issues pertaining to tropical agricultural research or related fields of study. It is published four times a year in February, May, August and November.

JST caters for science and engineering research or related fields of study. It is published twice a year in January and July.

JSSH deals in research or theories in social sciences and humanities research with a focus on emerging issues pertaining to the social and behavioural sciences as well as the humanities, particularly in the Asia Pacific region. It is published four times a year in March, June, September and December.

Why should you publish in Pertanika Journals?

Benefits to Authors

PROFILE: Our journals are circulated in large numbers all over Malaysia, and beyond in Southeast Asia. Recently, we have widened our circulation to other overseas countries as well. We will ensure that your work reaches the widest possible audience in print and online, through our wide publicity campaigns held frequently, and through our constantly developing electronic initiatives via Pertanika online submission system backed by Thomson Reuters.

QUALITY: Our journals’ reputation for quality is unsurpassed, ensuring that the originality, authority and accuracy of your work will be fully recognized. Each manuscript submitted to Pertanika undergoes a rigid originality check. Our double-blind peer reviewing procedure is fair and open, and we aim to help authors develop and improve their work. Pertanika JST is now over 35 years old; the accumulated knowledge has resulted in Pertanika being indexed in SCOPUS (Elsevier), EBSCO, DOAJ, CAB and AGRICOLA.

AUTHOR SERVICES: We provide a rapid response service to all our authors, with dedicated support staff for each journal, and a point of contact throughout the refereeing and production processes. Our aim is to ensure that the production process is as smooth as possible, is borne out by the high number of authors who publish with us again and again.

LAG TIME: Submissions are guaranteed to receive a decision within 14 weeks. The elapsed time from submission to publication for the articles averages 5-6 months. A decision of acceptance of a manuscript is reached in 3 to 4 months (average 14 weeks).

Call for Papers

Pertanika invites you to explore frontiers from all fields of science and technology to social sciences and humanities. You may contribute your scientific work for publishing in UPM’s hallmark journals either as a regular article, short communication, or a review article in our forthcoming issues. Papers submitted to this journal must contain original results and must not be submitted elsewhere while being evaluated for the Pertanika Journals.

Submissions in English should be accompanied by an abstract not exceeding 300 words. Your manuscript should be no more than 6,000 words or 10-12 printed pages, including notes and abstract.

Submissions should conform to the Pertanika style, which is available at www.pertanika.upm.edu.my or by mail or email upon request.

Papers should be double-spaced 12 point type (Times New Roman fonts preferred). The first page should include the title of the article but no author information. Page 2 should repeat the title of the article together with the names and contact information of the corresponding author as well as all the other authors. Page 3 should contain the title of the paper and abstract only. Page 4 and subsequent pages to have the text - Acknowledgments - References - Tables - Legends to figures – Figures, etc.

Questions regarding submissions should only be directed to the Executive Editor, Pertanika Journals.

Remember, Pertanika is the resource to support you in strengthening research and research management capacity.

An Award Winning International- Malaysian Journal

Feb, 2008

Mail your submissions to:
The Executive Editor
Pertanika Journals, UPM Press
Office of the DVC (R&D)
IDEA Tower II, UPM-MTDC Technology Centre
Universiti Putra Malaysia
43400 UPM, Serdang, Selangor
Malaysia
Tel: +6 03 8947 1622
nddepts@admin.upm.edu.my
www.pertanika.upm.edu.my