Change of Bioactive Constituent in *Clinacanthus nutans* Leaves under Sun Drying

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Abstract. *Clinacanthus nutans* (*C. nutans*) or locally known as belalai gajah is a folk medicine since ancient time. This research project was established to investigate the effects of under sun drying on the *C. nutans* bioactive constituent. The drying experiments were conducted using different drying surfaces i.e. perforated, black polythene and white polythene. The fresh and dried leaves were then extracted using a sonicator to evaluate its bioactive constituent. The total phenolic content (TPC) in the *C. nutans* extracts were determined using Follin Ciocalteu reagent method to represent the bioactive constituent. Drying over the white polythene surface showed the slowest reduction of moisture content as compared to the perforated polythene and black surfaces. Results also showed no significant effect of the drying surfaces on the TPC. However, the TPC in the dried leaves was significantly higher than in the fresh leaves. This may be due to the plant cells response to abiotic stress and the inhibition of oxidation enzymes. Therefore, drying *C. nutans* leaves under sun light could be considered in order to preserve the concentration of phenolic compounds and for minimizing energy consumption.

1. Introduction

*Clinacanthus nutans* (*C. nutans*) is a popular herbal plant in Malaysia and the neighbouring countries such as Thailand and Indonesia [1]. This plant has been used since ancient time in treating skin rashes, insect and snake bites, skin lesions caused by virus, diabetes mellitus, dengue fever and has diuretics properties [2]. Through a scientific investigation this plant has proved to exert anti-herpes simplex virus [3], antiproliferative effects on cultured human cancer cell lines [4], relief of skin inflammation and insect bites and varicella-zoster lesions [5].

*C. nutans* is a perishable herb due to its high water content (about 80%). Perishable materials are prone to microbial degradations so that it could not be stored in its fresh form. Drying technique is commonly used to preserve them during storage by reducing moisture level to a certain safe value. This preservation method is believed to maintain beneficial medicinal properties of *C. nutans* during storage promising a premium quality herbal raw material, as for the production of high quality nutraceutical products. Several drying techniques of both natural and artificial methods have been used to dry herbal materials to preserve their bioactive constituent such as under sun, shade drying, oven, microwave, freeze drying and vacuum oven. For instance, the microwave drying of parsley did not show any significant effect on its phenolic content compared to in the fresh leaves [6]. Another research work on *C. nutans* revealed the phenolic content reduced when the leaves were dried by freeze drying as compared to the oven and air drying [7]. This result showed that the bioactive content in *C. nutans* may not susceptible to heat. Thus, our research work is conducted to investigate the effect of natural sun drying on the total phenolic content of *C. nutans* leaves for complementing the existing studies. It is
important to establish the natural drying technique to our local herbs including *C. nutans* as to provide more options to the manufacturers especially in term of energy saving and maintaining bioactive constituent.

2. Material and methods

2.1. Sample preparation
The fresh *C. nutan* herbal plants were collected at the Institute of Sustainable Agrotechnology (INSAT), Universiti Malaysia Perlis (UniMAP). The uniform size leaves were sorted from the plant stems and rinsed with tap water. The leaves were kept in a fridge up to 48 hours prior to use.

2.2. Drying experiment
The fresh leaves were distributed on three different drying surfaces i.e. perforated, black polythene and white polythene. Each sample size was 30 g and the samples were prepared in three replications. The experiments were conducted under climatic condition of Campus UniCITI Alam, Universiti Malaysia Perlis, Perlis, Malaysia. The drying process was carried out during 1200 noon until 1700 afternoon. The weight of the sample was recorded every hour until it achieved saturation; weight change on the three consecutive readings was less than 0.001 g. The sample weights were found to become saturated within 3 days. The samples were kept in an air-tight container when not exposed to the sun light as to maintain their moisture content.

2.3. Moisture content determination
The *C. nutans* leaves were dried in an oven at 105°C, 24 hours to determine its dry matter weight (*m*<sub>dw</sub>). The moisture content (MC) was then calculated using an equation below:

\[
MC_{db} = \frac{m_t - m_{dw}}{m_{dw}}
\]  

where,
*MC*<sub>db</sub>= moisture content (dry basis)
*m*<sub>t</sub>=weight of the sample at time, *t*

The graph of leaves moisture content versus drying time was plotted.

2.4. *C. nutans* leaves extraction
The fresh and dried *C. nutans* leaves were ground using a laboratory blender. A 1.0 g sample of ground *C. nutans* was weighed and mixed with 100 ml distilled water. Then, the samples were extracted by placing it in ultrasonic bath for 30 min at the temperature of 40°C. The extracts were filtered using Wathman No. 4 filter paper. The supernatant was used for analysis of total phenolic content (TPC).

2.5. Total phenolic compounds determination
The TPC of *C. nutans* extracts were determined using Folin Ciocalteu reagent [8]. An amount of 0.5 ml leaves extract was mixed with 0.5 ml Folin Ciocalteu reagent. Then, 1.5 ml of sodium bicarbonate was added to the mixture. The mixture was then incubated in the dark for 30 minutes. The absorbance of the mixture was measured at 765 nm using a spectrophotometer. Distilled water was used as a blank solution. The concentration of phenolic compounds in the extracts were attained by comparison with a gallic acid calibration curve and expressed as mg GAE/g of extract.
2.6. Statistical analysis
The effects under sun drying using different drying surfaces on the TPC in the *C. nutans* leaves extracts were compared using the Analysis of Variance (ANOVA). Unless otherwise stated, the significance level was established at probability (P) < 0.05. For comparison between the means, the Student’s t-test was performed. Significance was established if the difference between the means for the treatments was more than the calculated least square difference (LSD).

3. Results and discussion
3.1. Relationship between moisture content and drying time
Figure 1 shows the reduction of moisture content versus time of *C. nutans* leaves under sun drying using different drying surfaces. Drying on the white surface showed the slower moisture content reduction than the other surfaces especially in day 2. It is because white surface does not absorb heat, drying process was only due to the natural heat from the sunlight. Perforated and black surfaces showed similar drying occurrence. Perforated surface allows air to reach entire leaf surface thus helped the drying process made faster while, black surface is known able to absorb heat thus accelerating the drying process.

As it can be seen in Fig. 1, the trend of moisture reduction was higher during day 2 due to high temperature was recorded. Although lower temperature was recorded during day 1, the drying rate was also driven by the higher moisture content in the leaves. During day 3, the removal of moisture content for all drying surfaces were getting slower which nearly equilibrium. The equilibrium moisture content determined was 0.18 g water/g dry basis to all drying surfaces.

![Figure 1](image)

**Figure 1.** Variation of the *C. nutans* leaves moisture content with time during sun drying.

3.2. Effect of sun-dried on total phenolic content (TPC) in *C. nutans* leaves extracts
The effect of TPC in the fresh and dried *C. nutans* leaves extracts is shown in Figure 2. The highest TPC in *C. nutans* was detected in the extracts under perforated drying surface (10.28 mg GAE/g), followed by black drying surface (9.695 mg GAE/g) and the white drying surface (9.380 mg GAE/g extract). These results showed that the effect of the drying treatments on TPC was not significantly different.
Figure 2. Effect of the drying surfaces on TPC in *C. nutans*.

The sun drying process significantly increased the TPC in the dried leaves extract as compared to that in the fresh leaves extract. This higher production of phenolic compound may be associated with a stress-induces responcse of the plant cells caused by the moisture loss during drying [9]. Drying also inhibits oxidative enzymes, which allowing better retention of phytochemicals in the dried leaves extracts [10]. In addition, the TPC in this sun-dried *C. nutans* leaves extracts is found higher than the leaves dried by oven, air and freeze drying [7]. Another research work showed that the TPC in the *C. nutans* also higher than in the sun dried misai kucing (*Orthosiphon staminus*) leaves [11]. Regardless of its disadvantages, under sun drying technique is appropriate to dry *C. nutans* herb due to the new findings revealed in this work. Therefore, drying of *C. nutans* herb under sun light could be considered in order to preserve bioactive constituent and minimize the operating cost.

4. Conclusion

From the results obtained, it can be concluded that under sun drying on the different drying surfaces employed in this work did not show any significant difference on the TPC. However it significantly increases the content of total phenolic compounds in the *C. nutans* leaves as comparison to in the fresh leaves. Thus, under sun drying method could be applied in order to improve the TPC content and as an option for energy saving processing method particularly on the *C. nutans* herb.

References

[1] Ariful A Sahena F Ghafoor K Hakim Juraimi A S Khatib A Zaidul I S 2016 A review of the medical pharmacologyphytochemistry, *Asian Pac J Trop Med* 9 402–409

[2] Goonasakaran S A 2013 Preliminary Antimicrobial and Phytochemical Analysis of *Clinacanthus nutans* and Azadiracht indica, Master Thesis Technological University of Malaysia, Johor, Malaysia

[3] Pongmuangmul S Phumiamorn S Sanguansermsri P Wongkattiya N Hamilton Fraser I Sanguansermsri D 2016 Anti-herpes simplex virus activities of monogalactosyl diglyceride and digalactosyldiglyceride from *Clinacanthus nutans*, a traditional Thai herbal medicine, *Asian Pac J Trop Biomed* 6 192–197

[4] Yong Y K Tan J J The S S Hui Mah S Chiong H S and Ahmad Z 2013 *Clinacanthus nutans* Extracts Are Antioxidant with Antiproliferative Effect on Human Cancer Cell Lines, *Evidence Based Complementary and Alternative Med* 1 1-8

[5] Charuwichitratana S Wongrattanapasson N Timpatanapong P Bunjob M 1996 Herpes zoster: treatment with *Clinacanthus nutans* cream, *Int J Dermatol* 35 665-666
[6] Magdalena S’ledz’ Malgorzata Nowacka Artur Wiktor Dorota Witrowa-Rajchert 2013 Selected chemical and physico-chemical properties of microwave- convective dried herbs, *Food and Bioprod Proc* 91 421–428

[7] Leng W K Ahmed Mediani Z Nur Khaleeda Zulaikha Sze W L Intan Safinar I Alfi Khatib Khozirah S Faridah A 2015 Phytochemical diversity of *Clinacanthus nutans* extracts and their bioactivity correlations elucidated by NMR based metabolomics *Phytochem Letters* 14 123–133

[8] Chong C H Law C L Luqman Chuah A Wan Ramli W D 2009 Drying Models and Quality Analysis of Sun-Dried Ciku *Drying Tech* 27 985–992

[9] Hossain M Barry-Ryan C Martin-Diana A B Brunton N 2010 Effect of drying method on the antioxidant capacity of six Lamiaceae herbs *Food Chem* 123, 85–91

[10] Hong-fang J Ai-lin D Ling-wen Z Chun-yang X Ming-duo Y Fang-fang L 2012 Effects of drying methods on antioxidant properties in *Robinia pseudoacacia* L. flowers, *J Med Plants Res* 6, 3233–3239

[11] Abdullah S Shaari A R Rukunudin I H Ahmad M S 2012 Effect of drying methods on metabolites composition of misai kucing (*Orthosiphon stamineus*) leaves *Procedia APCBEE* 178-182