ESTABLISHMENT OF OIL PALM SUSPENSION CULTURE IN THE
SIXFORS BIOREACTOR

By

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Thesis Submitted to the School of Graduate Studies, Universiti Putra
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Especially dedicated to all MMBPP’ ist....................
Abstract of the thesis presented to the Senate of University Putra Malaysia in fulfillment of the requirement for the Degree of Master of Science

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In vitro propagation is an important part of the oil palm industry’s approach towards clonal propagation of high-yielding materials. Oil palm suspension cultures have been established using the shake flask system which was developed for production of a reliable supply of regenerable plant tissues. However, this system is inefficient for fast large scale proliferation of embryogenic suspension cultures.

Bioreactors have been used for the industrial production of microbial, animal and plant metabolites. However, it’s used was not well known in oil palm suspension culture. During the development of oil palm suspension culture in the sixfors multibioreactor, nutrients and extra cellular metabolites were monitored where kinetic parameters and nutrients to biomass conversion yield were calculated to better characterise the behaviour of oil palm suspension culture. It was observed that the amount of biomass of all the cell lines was at an average of 2-3 fold higher than the original inoculums weight with an incubation period of 30 to 60 days.
The carbon source, which is sucrose, was hydrolysed to glucose and fructose in the first 10 days and both were completely utilised after the 25th day. The sugar to biomass conversion yield was low and the mainly linear growth showed that the growth of the cell was limited by the culture conditions. Nitrogen sources from the MS media remained in excess until the end of the growth period where only 30% of ammonia and 15% of nitrates were utilised which resulted in the cell being toxic and thus limiting cell growth.

The growth was exponential in the first 10 days with a maximum specific growth rate of 0.07 day\(^{-1}\) which corresponded to a doubling time of 10 days. The cells then entered a period of linear growth until Day 25 to reach the maximum dry weight of 4 g/l, after which the cells began to die off causing the dry weight to fall to 2.8 g/l at Day 45.

The pH profile was an indication of the nitrogen and sugar uptake by the cells. The pH decreased rapidly from 5.6 to 4.0 in the first 9 days and then increased gradually to 4.4 at the 25th day. At this point, the cell growth had stagnated, and the pH quickly increased to 5.5 before declining again to the end of the culture at Day 45. The initial pH decrease was partly due to the uptake of ammonium. After this, however, the great increase was due to the uptake of nitrate ions to the ammonium stored in the vacuoles of the cell.
This study provides a better understanding of oil palm suspension culture in a bioreactor with regards to the growth, nutrient uptake and metabolite production. This information will further enhance the progress of oil palm clonal materials development towards mass propagation production.
PEMBIAKKAN KULTUR AMPAIAN SAWIT DI DALAM BIOREAKTOR SIXFOR

Oleh

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Pembiakkan secara in vitro adalah kaedah utama industri sawit kearah pengeluaran klon yang mempunyai hasil yang tinggi. Kultur ampaian sawit telah berjaya dilaksanakan dengan menggunakan sistem kelalang goncang di mana ianya dilakukan secara berterusan untuk penghasilan anak klon sawit. Walaubagaimanapun, sistem ini kurang efisen untuk pengeluaran berskala besar kultur ampaian sawit embriogenik.

Bioreaktor telah digunakan didalam industri pengeluaran bagi sektor mikrobiologi, haiwan dan metabolit tumbuhan. Walaubagaimanapun, ianya jarang dilaksanakan pada kultur ampaian sawit. Di dalam kajian menggunakan bioreaktor sixfor, nutrien dan pengeluaran metabolit telah diselidik. Di samping itu parameter kinetik dan penggunaan nutrien berbanding dengan penghasilan biomass telah dikira untuk menggambarkan keadaan sebenar kultur ampaian sawit. Semasa proses inkubasi 30 ke 60
hari, biomass telah meningkat secara purata sebanyak 2-3 kali lebih tinggi daripada berat asal.


Pertumbuhannya di hari kesepuluhan adalah pada tahap eksponensial dengan kadar pertumbuhan spesifik sebanyak 0.07/hari yang berkadar sama dengan pengandaan sebanyak 10 hari. Sel kemudian melalui fasa pertumbuhan linear sehingga hari ke 25 untuk sampai ke tahap berat kering maksima 4 g/l. Ianya kemudian menurun ke 2.8 g/l di hari ke 45 bilamana sel mula mati.

Profil pH menunjukkan tahap pemakanan sumber nitrogen dan gula.. Di hari yang ke sembilan, pH menurun secara drastik dari 5.6 ke 4.0 dan
mengingkat secara perlahan sehingga hari ke 25 kepada 4.4. Di masa ini pertumbuhan sel terbantut serta kemudiannya pH menaik secara drastic ke 5.5 sebelum menurun balik diperlangkat pengakhiran kultur dihari ke 45. Penurunan pH dipemulaan adalah sebahagiannya disebabkan pemakanan ammonia. Selepas itu, peningkatan pH yang tinggi adalah disebabkan pemakanan nitrat ion ke pada ammonia yang disimpan didalam sel vakuol.

Kajian ini telah memberikan penambahan pengetahuan terhadap kultur ampaian sawit dari segi pertumbuhan, pengambilan nutrien dan pengeluaran metabolit. Bagi tujuan pengkomersialan, adalah diharapkan segala hasil kajian yang didapati akan dapat mempercepatkan proses pengeluaran klon sawit secara besar-besaran.
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I certify that an Examination Committee has met on 13th July 2006 to conduct the final examination of Samsul Kamal Bin Rosli on his Master of Science thesis entitled "Establishment of Oil Palm Suspension Culture in the Sixfors Bioreactor" in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulations 1981. The Committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follows:

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DECLARATION

I hereby declare that the thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.

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Date:
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