

ฤทธิ์ก่อกลายพันธุ์และฤทธิ์ต้านก่อกลายพันธุ์ของสารสกัดกวาวเครือขาว  
*Pueraria mirifica* กวาวเครือแดง *Butea superba* และกวาวเครือดำ  
*Mucuna collettii* ด้วยวิธีทดสอบแบบเอมส์

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**MUTAGENIC AND ANTIMUTAGENIC ACTIVITIES OF  
EXTRACTS FROM WHITE KWAO KRUA *Pueraria mirifica*,  
RED KWAO KRUA *Butea superba* AND BLACK KWAO KRUA  
*Mucuna collettii* BY AMES TEST**

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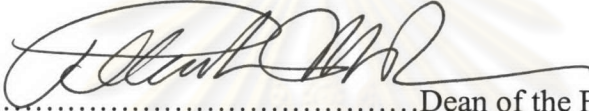
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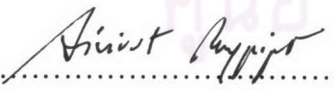
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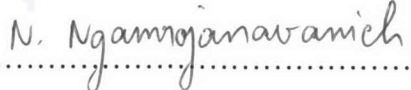
  
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เกศ พูลเจริญ: ฤทธิ์ก่อกลายพันธุ์และฤทธิ์ต้านก่อกลายพันธุ์ของสารสกัดกวางเครือขาว *Pueraria mirifica* กวางเครือแดง *Butea superba* และกวางเครือดำ *Mucuna collettii* ด้วยวิธีทดสอบแบบแอมส์ (MUTAGENIC AND ANTIMUTAGENIC ACTIVITIES OF EXTRACTS FROM WHITE KWAO KRUA *Pueraria mirifica*, RED KWAO KRUA *Butea superba* AND BLACK KWAO KRUA *Mucuna collettii* BY AMES TEST)

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ทดสอบความเป็นพิษต่อ *Salmonella* Typhimurium TA98 และTA100 ของสารสกัดจากกวางเครือทั้ง 3 ชนิด พบว่าสารสกัดจากกวางเครือดำ มีความเป็นพิษมากที่สุด ต่อ *S. Typhimurium* TA98 ผลการทดสอบฤทธิ์ก่อกลายพันธุ์ และฤทธิ์ต้านก่อกลายพันธุ์ ด้วยวิธีทดสอบแบบแอมส์ ที่ ระดับความเข้มข้น 0.625, 1.25 และ 2.5 mg/plate (100  $\mu$ l) ด้วย *S. Typhimurium* TA98 และTA100 ในภาวะที่มีและไม่มีเอนไซม์ S9 พบว่าสารสกัดจากกวางเครือขาว 28 จังหวัด กวางเครือแดง 24 จังหวัด และกวางเครือดำ 4 จังหวัด ไม่มีฤทธิ์ก่อกลายพันธุ์ ยกเว้นสารสกัดกวางเครือแดงจากจังหวัดราชบุรีเมื่อนำมาทดสอบด้วย *S. Typhimurium* TA98 ที่ความเข้มข้น 2.5 mg/plate (100  $\mu$ l) ในภาวะที่ไม่มีเอนไซม์ S9 ซึ่งมีความแตกต่างจากกลุ่มควบคุมอย่างมีนัยสำคัญ ( $P < 0.05$ ) การทดสอบฤทธิ์ต้านก่อกลายพันธุ์ที่ความเข้มข้น 2.5 mg/plate (100  $\mu$ l) ต่อสารก่อกลายพันธุ์ 2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide (AF-2) 0.01  $\mu$ g/plate (100  $\mu$ l) ด้วย *S. Typhimurium* TA 100 ในภาวะที่ไม่มีเอนไซม์ S9 พบว่าเปอร์เซ็นต์ยับยั้งการก่อกลายพันธุ์ด้วยสาร AF-2 ในกวางเครือขาวจากจังหวัดอุทัยธานี กวางเครือแดงจากจังหวัดกาญจนบุรี และกวางเครือดำจากจังหวัดเชียงใหม่ มีค่าเท่ากับ 25.75, 53.36 และ 50.55 ตามลำดับ เมื่อทดสอบด้วยสารก่อกลายพันธุ์ Benzo(a)Pyrene (B(a)P) 5 mg/plate ในภาวะที่มีเอนไซม์ S9 พบว่า กวางเครือขาวจากจังหวัดสุโขทัย กวางเครือแดงจากจังหวัดชลบุรี และกวางเครือดำจากจังหวัดกาญจนบุรี สามารถยับยั้งการก่อกลายพันธุ์ด้วยสาร B(a)P เท่ากับ 55.95, 88.61 และ 92.87 เปอร์เซ็นต์ตามลำดับ และจากการยืนยัน ฤทธิ์ก่อกลายพันธุ์ และฤทธิ์ต้านก่อกลายพันธุ์ ที่ ระดับความเข้มข้น 2.5, 5 และ 10 mg/well (10  $\mu$ l) เมื่อทดสอบด้วย *Bacillus subtilis* H17 and H45 ในภาวะที่ไม่มีเอนไซม์ S9 พบว่าสารสกัดกวางเครือทั้งหมด ไม่มีฤทธิ์ก่อกลายพันธุ์ และตรวจพบสารสกัดกวางเครือดำจากจังหวัดเชียงราย กวางเครือแดงจากจังหวัดเลย ที่ระดับความเข้มข้น 2.5 mg/well สามารถยับยั้งการก่อกลายพันธุ์ของสาร AF-2 0.1  $\mu$ g/well (10  $\mu$ l) อย่างมีนัยสำคัญ ( $P < 0.05$ ) และกวางเครือขาวจากจังหวัดอุทัยธานี ที่ระดับความเข้มข้น 0.1 mg/well สามารถยับยั้ง การก่อกลายพันธุ์ของสาร AF-2 อย่างมีนัยสำคัญ ( $P < 0.05$ ) โดยสรุปไม่พบฤทธิ์ก่อกลายพันธุ์ของสารสกัดจากกวางเครือทั้งหมด แต่พบฤทธิ์ต้านก่อกลายพันธุ์ในกวางเครือบางตัวอย่าง และพบฤทธิ์ต้านก่อกลายพันธุ์แรงที่สุดจากกวางเครือดำ

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ปีการศึกษา.....2548.....ลายมือชื่ออาจารย์ที่ปรึกษา.....

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KADE PULCHAROEN: MUTAGENIC AND ANTIMUTAGENIC ACTIVITIES OF EXTRACTS FROM WHITE KWAO KRUA *Pueraria mirifica*, RED KWAO KRUA *Butea superba* AND BLACK KWAO KRUA *Mucuna collettii* BY AMES TEST. THESIS ADVISOR: ASSOC. PROF. WICHAI CHERDSHEWASART, D.Sc., THESIS CO-ADVISOR: ASSOC. PROF. SIRIRAT RENGPIPAT, Ph.D. 153 pp. ISBN 974-14-2293-8

Cytotoxicity evaluation of the 3 plant extracts on *Salmonella* Typhimurium TA98 and TA 100 revealed that *Mucuna collettii* extract exhibited the strongest cytotoxicity on *S. Typhimurium* TA98. Mutagenic and antimutagenic activities of *Pueraria mirifica*, *Butea superba* and *Mucuna collettii* collected from 28, 24 and 4 provinces, respectively were tested. Their extracts were evaluated by Ames test using *S. Typhimurium* strain TA98 and TA100 under the absence and presence of metabolic activity conditions at the concentration of 0.625, 1.25 and 2.5 mg/plate (100  $\mu$ l) in comparison with the control. No mutagenicity was induced by all plant extracts except *B. superba* collected from Ratchaburi province which exhibited mutagenicity ( $P < 0.05$ ) in *S. Typhimurium* strain TA98 under the absence of metabolic activity condition at the concentration of 2.5 mg/plate (100  $\mu$ l). In the test with mutagenicity on *Salmonella* strains, all plant extracts exhibited significant inhibition ( $P < 0.05$ ) against 2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide (AF-2) and Benzo(a)Pyrene (B(a)P). *P. mirifica* collected from Uthai Thani, *B. superba* collected from Kanchanaburi, *M. collettii* collected from Chiang Mai inhibited mutagenicity induced by AF-2 0.01  $\mu$ g/plate (100  $\mu$ l) on *S. Typhimurium* strain TA100 at the inhibition percentage of 25.72, 53.36 and 50.55, respectively. *P. mirifica* collected from Sukhothai, *B. superba* collected from Chonburi, *M. collettii* collected from Kanchanaburi inhibited mutagenicity induced by B(a)P 5  $\mu$ g/plate on *S. Typhimurium* strain TA100 at the inhibition percentage of 55.95, 88.61 and 92.87, respectively. The mutagenicity and antimutagenicity assay of the plant extracts were confirmed in *rec* assay with *Bacillus subtilis* strain H17 and H45 under the absence of metabolic activity conditions. All plant extracts at the concentration of 2.5, 5 and 10 mg/well (10  $\mu$ l) exhibited no mutagenicity. *Mucuna collettii* collected from Chiang Rai and *Butea superba* collected from Loei exhibited significant inhibition ( $P < 0.05$ ) against AF-2 0.1  $\mu$ g/well (10  $\mu$ l) at the concentration of 2.50 mg/well (10  $\mu$ l), and *P. mirifica* collected from Uthai Thani exhibited significant inhibition ( $P < 0.05$ ) against AF-2 at the concentration of 10 mg/well (10  $\mu$ l). In summary, all Kwao Krua in this study showed no mutagenicity but antimutagenicity was detected in certain samples. The strongest antimutagenicity was found in *M. collettii*.

Field of study...Biotechnology .....Student's signature.....*Kade Pulcharoen*.....

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## LIST OF ABBREVIATION

°C	Degree Celsius
AF-2	2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide
B(a)P	Benzo(a)Pyrene
BW	Body weight
°C	Degree Celsius
cm	Centrimetre
DMSO	Dimethyl sulphoxide
DNA	Deoxy ribonucleic acid
hr	hour
IC <sub>50</sub>	Median Inhibition Concentration
Kg	Kilogram
L	Liter
LD <sub>50</sub>	Median Lethal Dose
LC <sub>50</sub>	Median Lethal Concentration
MGA	Minimal Glucose Agar
MW.	Molecular Weight
m	Metre
mm	Millimetre
μM	Micromolor
μg	Microgram
μl	Microliter
ml	Milliliter
min	Minute
ng	Nanogram
PI	Percentage Inhibition
S.E.M.	Standard Mean Error