

行政院所屬各機關因公出國人員出國報告書
(出國類別：出席國際會議)

出席第二十四屆國際蔗糖技術學會論文發表會報告

服 務 機 關：台灣糖業股份有限公司

出國人姓名職稱：鄭鴻財（總經理室總經理）

王隆輝（台糖研究所所長）

劉禮恭（糖業處處長）

張翊袖（台糖研究所園藝系主任）

黃啟民（台糖研究所糖業系主任）

劉嘉哲（台糖研究所產品開發系副研究員）

出國地點：澳洲布里斯本市

出國期間：九十年九月十四日至九月二十二日

報告日期：九十年十一月十二日

行政院研考會/省(市)研考會 編號欄
F4 / C09005907

行政院及所屬各機關出國報告提要

頁數: 48 含附件:是

報告名稱:

出席第二十四屆國際蔗糖技術學會論文發表會報告

主辦機關:

台灣糖業股份有限公司

聯絡人/電話:

黃啟民/06-2671911-431

出國人員:

鄭鴻財	台糖公司	總經理室	總經理
王隆輝	台糖公司	研究所	所長
劉禮恭	台糖公司	糖業處	處長
張翊袖	台糖公司	研究所	系主任
黃啟民	台糖公司	研究所	系主任
劉嘉哲	台糖公司	研究所	副研究員

出國類別: 其他(出席國際會議)

出國地區: 澳洲

出國期間: 民國 90 年 9 月 14 日-90 年 9 月 22 日

報告日期: 民國 90 年 11 月 12 日

分類號/目: / /

關鍵詞:

內容摘要:

- 一、國際蔗糖技術協會(International Society Of Sugar Cane Technologists), 為一蔗糖製造業者之世界性組織。該協會每三年輪流在世界各主要產糖國家召開論文發表會, 台灣糖業公司每屆依例派代表出席與會及宣讀論文, 達到學術交流目的, 並學習各先進國家之甘蔗育種、栽培、副產品及製糖等新技術。本年度為第二十四屆, 由鄭鴻財總經理領隊及王隆輝所長率團出席, 由台糖研究所劉嘉哲主任宣讀兩篇論文, 另由黃啟民主任發表兩篇海報論文, 題目分別為(1)利用微生物從蔗糖漿生產果寡糖。(2)含硒酵母的生產。(3)製糖工場碳酸法程序生產金二砂製程的建立。(4)廢蒸汽回收的自動控制系統。
- 二、宣揚我國製糖技術及副產品等研發成果, 引起國際重視, 提升我國國際地位。觀摩其他產糖國家的長處, 彌補我國之短處。藉此與國際製糖人士交往, 增加彼此聯絡管道, 並可開發海外投資之商機。

本文電子檔已上傳至出國報告資訊網

行政院及所屬各機關出國報告審核表

出國報告名稱：出席第二十四屆國際蔗糖技學會論文發表會報告	
出國計畫主辦機關名稱：台灣糖業股份有限公司	
出國人姓名/職稱/服務單位：鄭鴻財等六人/總經理/總經理室	
出國計畫主辦機關審核意見	<input type="checkbox"/> 1. 依限繳交出國報告 <input type="checkbox"/> 2. 格式完整 <input type="checkbox"/> 3. 內容充實完備 <input type="checkbox"/> 4. 建議具參考價值 <input type="checkbox"/> 5. 送本機關參考或研辦 <input type="checkbox"/> 6. 送上級機關參考 <input type="checkbox"/> 7. 退回補正，原因： <input type="checkbox"/> ①不符原核定出國計畫 <input type="checkbox"/> ②以外文撰寫或僅以所蒐集外文資料為內容 <input type="checkbox"/> ③內容空洞簡略 <input type="checkbox"/> ④未依行政院所屬各機關出國報告規格辦理 <input type="checkbox"/> ⑤未於資訊網登錄提要資料及傳送出國報告電子檔 <input type="checkbox"/> 8. 其他處理意見：
層轉機關審核意見	<input type="checkbox"/> 同意主辦機關審核意見 <input type="checkbox"/> 全部 <input type="checkbox"/> 部分_____（填寫審核意見編號） <input type="checkbox"/> 退回補正，原因：_____（填寫審核意見編號） <input type="checkbox"/> 其他處理意見：

說明：

- 一、出國計畫主辦機關即層轉機關時，不需填寫「層轉機關審核意見」。
- 二、各機關可依需要自行增列審核項目內容，出國報告審核完畢本表請自行保存。
- 三、審核作業應於出國報告提出後二個月內完成。

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一、目的

(一)、國際蔗糖技術協會(International Society Of Sugar Cane Technologists)，為一蔗糖製造業者之世界性組織。該協會每三年輪流在世界各主要產糖國家召開論文發表會，台灣糖業公司每屆依例派代表出席與會及宣讀論文，達到學術交流目的，並學習各先進國家之甘蔗育種、栽培、副產品及製糖等新技術。本年度為第二十四屆，由鄭鴻財總經理領隊與王隆輝所長率團出席，由台糖研究所劉嘉哲代主任宣讀兩篇口頭論文，另由黃啟民主任發表兩篇海報論文，題目分別為(1)利用微生物從蔗糖漿生產果寡糖。(2)含硒酵母的生產。(3)製糖工場碳酸法程序生產金二砂製程的建立。(4)廢蒸汽回收的自動控制系統。

(二)、宣揚我國製糖技術及副產品等研發成果，引起國際重視，提升我國國際地位。觀摩其他產糖國家的長處，彌補我國之短處。藉此與國際製糖人士交往，增加彼此聯絡管道，並可開發海外投資之商機。

二、出國行程

90.09.14	台南→高雄→台北→澳洲布里斯本(啟程及轉機)
90.09.15	預備會議
90.09.16	報到及參加歡迎酒會
90.09.17	大會及論文發表會
90.09.18	論文發表會
90.09.19	論文發表會
90.09.20	論文發表會
90.09.21	論文發表會
90.09.22	澳洲布里斯本→台北→高雄→台南(回程)

三、參加國際蔗糖技術學會心得及建議

(一) 概述

國際蔗糖技術學會成立於1924年，至今已有76年的歷史，大約每隔3年舉辦一次會議。今年為第24屆，且為21世紀的第一次會議，意義重大。此會議網羅大學、公司及工廠的學者、專家和技術人員，共聚一堂，互相觀摩，主要目的是鼓勵創新的研發及發展經濟可行的新技術，透過成果分享的方式，提高製糖工業的利潤，使其能永續經營。澳洲為產糖大國，種蔗面積達52萬公頃，糖年產量約為400萬公噸。澳洲共有29座糖廠，其中26座位於昆士蘭省，而該省生產的粗砂，約佔澳洲全國產量的95%。澳洲亦為糖的出口大國，年營業額高達10億美金，外銷85%的粗砂至15個國家。另外，每年亦外銷26萬公噸的白糖，其精煉白糖的能力更高達每年130萬公噸。此外，該國亦擁有容量200萬公噸的大倉儲，更提升了調節供應的競爭力。澳洲曾在1935及1950年主辦過兩屆ISSCT大會，今年為第3次主辦，由於主辦國有雄心籌辦一次高標準及高效率的會議，豎立典範。在會場地點的選擇及軟硬體設備的安排等，都令人非常滿意。另外又提供免費渡船及公車作為與會人員的交通工具，更令人印象深刻。而印刷精美的論文集亦在報到時同步發放，可說是效率一流，令人欽佩。

(二) 出席情形

本屆出席會員共有728名，出席國家有法國、德國、印尼、瑞典、丹麥、比利時、荷蘭、義大利、委內瑞拉、阿拉伯聯合大公國、伊朗、印度、澳洲、美國、加拿大、泰國、墨西哥、巴西、英國、斐濟、馬特尼克、模里西斯、南非、瓜地馬拉、菲律賓、哥倫比亞、多明尼加、辛巴威、厄瓜多、肯亞、秘魯、中國、巴貝多、阿根廷、西印度、巴基斯坦、巴布新幾內亞、日本、越南、坦尚尼亞、哥斯達尼加、尼加拉瓜、新加坡、以色列、香港與中華民國等46國。

(三) 論文發表

台糖研究所糖業系主任黃啟民及產品開發系代主任劉嘉哲代表本公司，於本屆大會分別發表兩篇海報論文及口頭論文(詳見附錄)，題目分別為：

- A. 製糖工場碳酸法程序生產金二砂製程的建立
Process Modification for the Production of Golden B-Grade White Crystal Sugar
- B. 廢蒸汽回收的自動控制系統
An Automatic Control System for Recovering Exhaust Steam
- C. 利用微生物從蔗糖漿生產果寡糖
Production of Fructooligosaccharides from Sucrose Syrup by A Microorganism TSC-FOS1
- D. 含硒酵母的生產
Production of Selenium Yeast

(四) 大會論文摘要

本屆論文宣讀於90年9月17日上午10時，由國際蔗糖技術協會秘書及財務長Brian Egan等貴賓先後祝辭，歡迎世界各國與會代表參加本屆年會及論文發表大會後，正式展開為期五天之論文發表大會。

本屆農業組論文分為農藝、環保、農機、分子生物、昆蟲、植病及育種等項，共有61篇口頭論文及51篇壁報論文。工業組論文包括壓榨、清淨、過濾、蒸發、結晶、能源及廢棄物利用等項，共有25篇口頭論文及22篇壁報論文。副產品組包括健康食品、氨基酸測定、生物可分解塑膠、酒精生產及脫色等項，共有11篇口頭論文。另外也舉辦兩場專題研討會，主題分別為：(A)從採收到包裝過程中蔗糖的損失(B)分子生物技術於蔗糖生產之應用。

論文發表的工具，大部份皆使用手提電腦配合三槍投影來發表，內容相當生動且多彩多姿，但在宣讀時，仍有發生電腦當機而不知如何操作的窘況，未來若準備以此種設備作為論文宣讀工具，必需將電腦操作的功夫練好，才能避免當場出糗。以下為三十篇精選論文之摘要說明：

**MEMBRANE FILTRATION TECHNOLOGY IN THE CANE
SUGAR INDUSTRY**

薄膜過濾技術在製糖工業的角色

R.T.Steindal

Sugar Research Institute, Mackay, Queensland, Australia

雖然薄膜技術幾乎曾被所有的產蔗國家評估其可能帶來的潛在利益，但此技術目前仍未被製糖工場廣泛採用。此篇文章審視薄膜技術發展的現況，考慮導引製糖工業逐漸採用此技術。首先將報導薄膜過濾技術對製糖工業帶來的好處，而正在進行的研究及發展中的專利製程，有許多是透過薄膜技術作為工具，來提升離子交換及色層分析技術的分離效率，而使粗砂工場能夠直接生產白糖。同時，亦將討論阻隔液中糖損失及其減少損失的方法。使評估選擇適當處理阻隔液的方法，較易進行。

**CHROMATOGRAPHIC APPLICATIONS IN THE CANE
SUGAR INDUSTRY**

層析技術在蔗糖工業上的應用

Mike Kearney and Vadim Kochergin

Amalgamated Research Inc., Twin Fall, Idaho, USA

用來處理含糖溶液的工業級層析技術，目前已經是成熟且可靠的。在美國，超過 90% 以上的甜菜糖蜜是經由使用層析技術的方法來處理的。美國結晶糖業公司 (American Crystal Sugar Co.) 在北達克達州 (North Dakota) Hillsboro 的工廠，擁有最大的設施，每天處理量達 545 公噸 (80% 的乾燥糖蜜)。另外，兩座屬於 Amalgamated Sugar Co. 的糖廠，每座每天的處理量亦達 455 公噸。由這些工廠以層析技術來分離純化蔗糖效率頗高的成功經驗來看，未來利用此技術於甘蔗糖漿的清靜製程，似乎是無可避免的。最近許多新的發明包括利用 Fractal Technology 技術來縮減工業級層析系統設備的尺寸等，將可大幅降低成本。因此，未來層析技術廣泛應用於蔗糖工業的美夢，將可成真。

**THE USE OF BIG/GT TECHNOLOGY IN SUGAR
MILLS**

糖廠使用 BIG/GT 技術發電的經驗

F.A.B.Linero, H.M.Lamonica, and M.R.L.V.Leal

Centro de Tecnologia Copersugar, Sao Paulo, Brazil

由於巴西政府通過了新的環境政策，而使收穫前焚燒甘蔗的方法逐漸式微。因此，增加了人們對研發甘蔗夾雜物利用替代方案的興趣。其中一項具挑戰性的方案，就是以夾雜物為燃料來產生電熱能。而以生質為材料，利用生質合成式燃氣渦輪機 (BIG/GT) 產生電熱能是較先進的技術，此方法可較凝結/抽汽式蒸汽渦輪機 (CEST) 的方式，使每公噸甘蔗產生更多的電熱能。本文將介紹有關的汽化技術 (Gasfication Tech.)，目前在各領域的進展。並且提出一個 BIG/GT 真實的例子，介紹有關一座典型的巴西糖廠，如何利用 BIG/GT 產生電熱能，並將電能外售的實例，另外，也會介紹該糖廠如何有效利用夾雜物與蔗渣的方法，並分析其投資成本。

**OPPORTUNITIES FOR IMPROVING THE
MANAGEMENT OF SUGARCANE PRODUCTION
THROUGH THE ADOPTION OF PRECISION
AGRICULTURE-AN AUSTRALIAN PERSPECTIVE**

採用精準農業來改善甘蔗經營的機會

R.G.V. Bramley and THE LATE R.P. Quabba

CSIRO Land and Water, Australia

精準農業 (PA) 是個包羅萬象的名詞，常出現在許多可促進提升農業生產及經營的技術上，而技術的提升乃經由對農業土地生產潛能多變化的體認，即使距離很近，甚至只有幾公尺，生產力仍有變化。精準農業的核心技術包括產量監控 (Yield Monitors)，衛星定位系統 (Global Positioning System) 及地理資訊系統 (Geographical Information System)。本文將參考精準農業哲理目前在其他工業應用的現況，提出對精準農業未來的看法。其中將回顧近兩年來，實施精準農業對於澳洲 Herbert River 地區甘蔗產量的影響，亦描繪最近精準農業應用於葡萄酒生產的研究現況，更從中確認出，經由採用精準農業而能改善甘蔗產量及管理良方的機會。總之，在澳洲利用精準農業來控制蔗糖生產是理想可行的，但為了獲得成功，許多必要的操作改變，特別是收穫管理是無法避免的。

**THE ANALYSIS OF DEXTRAN AND SARKARAN IN
CANE PRODUCTS USING ENZYMIC HYDROLYSIS AND
HPAEC**

**利用水解酵素及 HPAEC 分析蔗糖中的 DEXTRAN 和
SARKARAN**

P.G. Morel Du Boil

Sugar Milling Research Institute, Durban, South Africa

本文敘述各種甘蔗產物中，多醣類如 Dextran 和 Sarkaran 的分析方法。分析的步驟是先分離出此高分子物質，接著再選擇利用適當的工業級水解酵素，來水解各別的多醣類。有特性的寡糖產物是以 High Performance Anion Exchange Chromatography With Pulsed Amperometric Detection (HPAEC-PAD) 來分析。這些方法已被採用來監控蔗汁和蔗糖中多醣類的分佈量及其在季節性和地理性上的差異，同時亦監控環境因子是否對其也有影響。

**DIRECT PRODUCTION OF WHITE SUGAR IN CANE
MILLS: TECHNICAL AND ECONOMIC ASPECTS**
由粗砂製糖工場直接生產白糖：技術和經濟面

V. Kochergin, M. Kearney and J.F. Alvarez

Sugar Cane Growers Cooperative of Florida, USA

本文討論由粗砂廠直接生產白糖的進展，目前是由一個長期且連續的大型研究來評估新製程的可行性。大型試驗結果的資料顯示，除了生產白糖，新製程亦可生產一些特高品質的粗砂。濃縮清淨汁經由結晶程序而得之粗砂樣品，未經過洗糖程序，而直接經由薄膜過濾、軟化和層析等處理後，可有效降低成品糖的色值、轉化糖、混濁度和灰分。估計約能增加 6-7% 的回收率。文中亦分析新製程的總成本結構 (Capital Cost Structure) 及每一單元的相對貢獻。基於最近的發展，許多製程改善的方案也將摘要出來。

**EXPERIENCES WITH OXIDATIVE DECOLOURISING PROCESSES IN THE
SOUTHERN AFRICAN SUGAR CANE INDUSTRY**

南非製糖工業於氧化脫色製程的經驗

S. B. Davis

Sugar Milling Research Institute, Durban, South Africa

雖然目前在煉糖工業上很少使用，過氧化氫及臭氧確是兩種可用在煉糖脫色

的強氧化劑。本文介紹南非糖業研究所應用此兩種化學藥品於製糖工業上的十餘年經驗。在溶糖後碳酸飽和前，加入過氧化氫或臭氧處理，確實證明可有效地去除糖的色素和其前驅物。工廠試驗結果顯示，於100-200ppm (on Brix) 的添加量，對白糖的色值有很大的改善空間，未來將繼續進行更進一步的試驗。

ENERGY EFFICIENT PROCESS FOR SUPERIOR QUALITY SUGAR

以能源有效利用之製程來生產高品質白糖

Sunil Singhal, Anup Kesarwani and Mangal Singh

India

高品質白糖製程 (DSQSP) 是在印度發展出的新製程，可由甘蔗直接生產與精煉糖相等級的白糖。此製程是專為雙重亞硫酸法廠建立的，不但可提升糖的品質，且不需要再結晶。製程中最重要之考量就是能源的需求，超出糖廠運轉所需能源之節餘蔗渣，其實是有很高經濟價值的，可用來汽電共生、造紙或是當燃料出售。但成本的考量也是非常重要的，尤其是對雙重亞硫酸法廠，因製程改變而增加設備費用，或因藥品和消耗品等使用量提高而增加費用。新製程中最重要之單元，就是糖漿以磷酸浮選及其過濾系統，此系統已在兩個每天分別壓榨 11,000 和 7,000 公噸甘蔗的糖廠中運轉。另外，活性碳處理單元及連續過濾-脫糖-脫水技術單元都已在小型試驗測試過，未來在大型試驗成功的機會，頗為樂觀。

SUGARCANE TRASH RECOVERY FOR USE IN POWER GENERATION

利用甘蔗夾雜物發電

Suleiman J. Hassuani

Copersucar Technology Center, Piracicaba-SP, Brazil

蔗糖工場如設有高效率的汽電共生系統，是可利用蔗渣生產出超過工廠所需的電量，此多餘的電，則可外售處理。但目前的狀況是當甘蔗收穫完畢後，由於沒有蔗渣作為燃料，汽電共生系統也要跟著關閉。為了使非開工期也能繼續發電，替代性燃料是非常需要的。而甘蔗夾雜物正是一種有潛力且吸引人的替代燃料，甘蔗夾雜物普遍存在於蔗田，目前由於環境保護的觀念日益提升，故禁止甘蔗收穫前焚燒夾雜物，因此，夾雜物的適當出路亦急需尋求。本文敘述巴西糖業技術中心發展出的有關夾雜物回收的方法，及介紹詳細的夾雜物包裝回收系統。另外，也報告夾雜物的包裝操作容量、包捆參數及其回收與搬運至工場的成本。同時，亦說明夾雜物的回收對農藝成本的影響。

HANDLING OF CLARIFER MUDS: LOSS MANAGEMENT

強化清淨污泥處理來降低糖損失

B. St. C. Moor

St. Clair Consultants, Cowies Hill, South Africa

在傳統糖廠的製程中，蔗汁是以凝聚劑處理後，靜置沉澱來分離出懸浮固體 (Suspended Solid)，雖然污泥中大部份的糖，可於真空過濾機中沖洗回收，但仍有 0.1-3% 的總糖量損失是發生在此過濾步驟的。這裡的糖損失可分成兩種型式，其一為殘留在污泥餅中的糖份，另一為經常被忽略掉或低估的微生物破壞。本文將介紹減少這些損失的操作良方，且提供一些標準製程控制的指引 (Guideline)。最近的研究成果顯示，很有希望將清淨過濾製程的總糖損失減少 50-80%，而此新方法是將污泥餅回送至壓榨及抽取製程中處理。

NEW FARMING SYSTEMS FOR SUGARCANE PRODUCTION

甘蔗生產新式栽培系統

T. A. Bull, C. P. Norris, B. G. Robotham and M. V. Braunack

Bureau of Sugar Experiment Stations, Bundaberg, Queensland 4670, Australia

澳洲工業的機械化發展不僅使工作效率提高且對田間栽培作業產生很大的革命。但現行機械化制度亦產生不少缺點如明顯增加土壤的壓實，損害土壤的完整性，雜草控制不易加上採收搬運時造成作物損失及傷害植株。因此亟須發展一套新的田間作業系統。根據最新的研究顯示窄行距顯著增加產量，並且以經濟型的引擎即可在田間執行此項田間作業。試驗一個畦植雙行，三行，四行蔗的窄行距，均較傳統的單畦單行的行距有利。不但克服光線、水份的互相攔截問題且克服土壤壓實、雜草控制、土壤完整性、水份利用效率等問題，並可避免採收時作物的損失及植株的損傷。但窄行距以目前農機具無法進行田間作業，必須改良現行農具發展適合此新的田間作業系統的多行開溝器。

PROSPECTS OF APPLYING CANE RIPENERS THROUGH IRRIGATION WATERS : RIPENING RESPONSE OF DINITROSCIFROL AND TRIACONTANOL

利用灌溉水施用成熟劑 DINITROSCIFROL 和 TRIACOTANOL 之成熟反應

S. Solomon and H. N. Shahi

Indian Institute of Sugarcane Research, Lucknow-226 002, India

利用生物調節劑或成熟劑來提高甘蔗的糖份在世界各地甘蔗商業栽培已採用。利用人工促使甘蔗成熟已有相當成功的例子，如澳洲使用 CP41854，瓜地馬拉用 Dalapon 均試驗的很成功。潛在的甘蔗成熟劑包括 Polaris, Ethrel, Pesco1815, Fusilade Super 和 Embark 已在夏威夷，澳洲，模里西斯，波多黎哥，哥倫比亞和南非等國家商品化利用，其他像 Ethrel, Glyphosate, Dinitrosocifrol, 和 Triacontanol 等甘蔗成熟劑在印度已經做了試驗以期在開工初期促成甘蔗成熟。文獻顯示使用這類化學品促進甘蔗成熟結果並不一致，因為受到土壤中 N 狀態，作物成熟性，甘蔗品種等之影響。過去報告顯示增加糖份回收幅度自 0.2 到 1.0 單位不等。某些國家利用甘蔗成熟劑已成為甘蔗栽培的一部份，它們利用開花噴撒器和 micro-light+飛機將甘蔗成熟劑噴撒在蔗田。對小農制的印度而言，此法不可行，因此另想辦法以灌溉水使用新種類的成熟劑並且不會失去它們的生物活性。本文乃利用兩種化學藥品 Dinitrosocifrol (每公頃 10 公斤) 和 Triacontanol (每公頃有效成分 10 公斤) 來試驗，結果對甘蔗成熟有促進效果，糖度百分比增加 0.2 至 1.0 單位。並且對莖部品質和宿根未造成不良作用。

DEVELOPMENT OF EST-DERIVED RFLP MARKERS FOR SUGARCANE BREEDING

發展 EST-衍生的 RFLP 標記輔助甘蔗育種

Jorge A.G. Da Silva, Eugenio C. Ulian and Carla F. Barsalobres

Copersucar Technology Center, Brasil

分子標記是染色體組分析的一個很好工具，可用於基礎的和應用的研究。RFLP 標記首先由 Burquist 於 1991 年應用於甘蔗屬雜種的遺傳變異分析。RFLP 圖可以提供一個更直接方法去選拔與數量性基因座有關的標誌。最近數量性狀基因座利用異質體 DNA 作 RFLP 探討已在甘蔗野生種雜交被鑑定出來。

依據一個有機個體表現部份序列，其表現序列標牌—Est 可以鑑定不同表現組織的基因。最近一個甘蔗 Est 計劃—SUCEST—已經在 FAPESP (聖保羅州政府研究部門) 開始。SUCEST 資料庫已選出 44524 個基因，除了基因鑑定和表現的資料外，EST 資料庫代表 microsatellite 和 RFLP 分子標記的一個分子標記與已知基因有關連時可容許間接而正確的選拔理想基因，對甘蔗育種將是一個正面的衝擊。在巴西甘蔗育種計劃的商業品種中發展 Est-衍生的 RFLP 標記，可以標記糖分和抗病的數量性狀基因位置圖，並且 Est-衍生的 RFLP 可以標記蔗糖代謝和抗病基因的蛋白質，利用此標記作南方分析繪出商業基因型的基因圖。

DEVELOPING SPECIES-SPECIFIC DNA MARKERS TO ASSIST IN
SUGARCANE BREEDING

發展種間特定 DNA 標記輔助甘蔗育種

Y.-B. Pan¹, D.M. Burner² and Q.Wei¹

USDA-ARS, ¹Louisiana, ²Arizona, USA

本文報告研發了兩個種間特定的 PCR 標記，Eri3/Eri4 和 Gig I /P II，並用來成功地鑑定出種間雜種。Eri3/Eri4 標記的大小為 313bp 它乃單獨由 Erianthus (芒) 的雜種擴大來的。另一標記 Gig I /P II 的大小為 176bp 是由甘蔗屬 giganteum 三個細胞體 (染色體數分別為 30, 60 和 90) 和芒品系得來的大小為 350bp。分子增殖和定序用來揭示從芒品系得來的 313bp Eri3/Eri4 標記和 350bp Gig I /P II 標記的核糖核酸序列。利用此二個種間特定標記可在實生苗初期鑑定種間雜種。因為甘蔗種間雜交去雄不易，大多數未去雄，利用種間特定分子標記在實生苗階段鑑定真種間雜種對甘蔗種間育種效率有很大助益。

EVALUATION OF THE BIOLOGICAL CONTROL PROGRAM AGAINST
SUGAR CANE STALK BORER, DIATRAEA SACCHARALIS f., AFTER
TWO DECADES OF FIELD ESTABLISHMENT OF COTESIA FLAVIPES
(CAMERON) IN JAMAICA

評估二十年來以 COTESIA FLAVIPES 在牙買加生物防治甘蔗莖部
螟害 DIATRAEA SACCHARALIS 計畫之得失

Trevor Falloon

Sugar Industry Research Institute, Kendal Road, Mandeville,
Jamaica

牙買加蔗田自 1972-1974 及 1980-1987 利用引進的寄生昆蟲 Cotesia flavipes 在實驗室培養後釋放田間來防治甘蔗莖部螟害 Diatraea saccharalis。田間試驗在 1982 年正式成功，並將寄生昆蟲快速釋放讓其群居在大部份蔗田。自 1999 年 11 月至 2000 年 3 月由三個蔗田收集螟蟲，目前調查幾乎為零感染。但是整個蔗區採收的甘蔗進工廠壓榨仍有 7% 以上的節間發生螟害。在最後一次釋放此寄生昆蟲顯著的降低蔗莖的為害。調查顯示維持生物防治效果仍須擴大釋放此寄生昆蟲。在灌溉區的蔗田，C. flavipes 防治螟害的效果較不完整，此問題仍待解決。

AN EPIDEMIC OF ORANGE RUST ON SUGARCANE IN AUSTRALIA

澳洲黃銹病的疫病學

R.C.Magarey, B.J.Croft and T.G.Willcox

Bureau of Sugar Experiment Station, Australia

澳洲甘蔗品種 Q124 於 2000 年期在昆士蘭發生黃銹病疫病，黃銹病在澳洲已存在一百多年，但只在 1900 年代早期對高貴蔗產生明顯的損害。Q124 品種以前是抵抗黃銹病的品種，目前嚴重感染黃銹病懷疑是黃銹病的新變異種造成此次的疫病。Q124 品種佔昆士蘭甘蔗栽培面積 45%，2000 年期糖量的損失歸因於昆士蘭中部地區，Berdekim, Herbert 和此方地區的 Q124 感染嚴重的銹病所致，其感染率高達 30-40%，同時造成噸糖和糖分的損失。此病減少甘蔗纖維含量，工廠必須購買額外的燃料來操作鍋爐。其他感染品種包括 Q173, Q178 和 Q182 全部的損失估計達到澳幣 150-210 百萬，此乃歷年來造成糖業損失最大的病害疫病。Q124 在黃銹病爆發前，它的產量較其他地區的許多品種均優。然而，在 2000 年的品種試驗；Q124 因感染黃銹病產量大減。目前澳洲糖試所百分六十五的商業品種和百分之十以下的種原保存品系和試驗中品系感染此新型的黃銹病，選育出優良抗病品系才是唯一解決之道，目前已育出抗病的新商業品種外放種植，但是 Q124 在田間種植太多。須再 3 年的時間才能全部從田間移除。目前是以殺菌劑 Mancozeb, tebuconazole 和 propiconazole 在田間施用以減少經濟上的損失。

EFFECT OF SUGARCANE YELLOW LEAF VIRUS ON SUGARCANE

YIELD AND JUICE QUALITY

甘蔗黃葉毒素病對甘蔗產量和蔗汁品質的影響

M.P.Grisham¹, Y-B.Pan¹, B.L.Legendre², M.A.Godshall³ and G.Eggleston⁴

¹USDA-ARS,Houma; ²Louisiana State University; ³SPRINew Orleans;

⁴USDA-ARS,New Orleans, USA

1996 年路州甘蔗植株感染黃葉毒素病 (SCYLV)，據報引起黃葉病癥狀 (YLS)。田間試驗評估此病對路州甘蔗的潛在衝擊。第一組試驗，比較 LCP82-89 品種感染 SCYLV 與未感染之植株的產量和蔗汁品質。結果一宿和二宿感染的甘蔗單位面積的糖量分別減少 11% 和 14%，感病植株的莖數和蔗量亦減少，但是品質包括錘度，糖分，纖維和純度百分比在感染與未感染植株間並無顯著差異。第二組試驗，自 LCP82-89 和 LH082-153 品種的一宿和二宿的感染株和未感染株分析葉部和莖部葉汁品質。結果甘蔗品質組成份在品種間並無差異。從 SCYLV 感染的葉子取下的汁液，糖份% 在 LCP82-89 較高而純度%、澱粉濃度，兩個品種感染株均較未感染者高，

但是總多糖類和果寡糖類均未發現有差異。聚合糖含量不一定。採收時甘蔗以機械收穫梢頭部的綠葉將被除去，但是倒伏的甘蔗頭部綠葉將可能一併送進廠壓榨，在此狀況下綠葉含量較高的澱粉可能減少工廠效率。

STABILITY OF MORPHOLOGICAL MARKERS OVER LOCATIONS, FOR THE IDENTIFICATION OF SUGARCANE VARIETIES

形態標記鑑定甘蔗品種

S. Hartatik¹, A. Makmur², A. Saefuddin² and S. Lamadji³

¹Jember University; ²Bogor Institute of Agriculture; ³ISRI, Indonesia

鑑定甘蔗品種對甘蔗育種相當重要。傳統上，利用形態標記鑑定品種以記分方式來記錄此標記，且易建檔、易評估。

近年來，利用分子標記供育種使用或做遺傳工程或在某種程度上做為植物材料的鑑定之用。但利用分子標記在育種計劃上並不實用，因為須花費高額的經費去建立特殊的設備和維持昂貴的勞力，所以形態上的標記仍為育種者和分類學者用來區分和分類類似品種的最佳選擇。Guiard (1995) 報告 UPOV (國際保護品種聯盟) 使用外表型的描述來進行品種的分散度，均一度和穩定度 (DUS) 試驗。形態特性被認為是遺傳所控制包括上位基因交感或二個基因控制並且受制於基因型與環境的交感作用，形態標記可以做為評估和描述種原的一個可靠工具。以形態標記描述和分類若干作物如大豆和野生玫瑰與分子標記如 RAPDs 非常吻合。在甘蔗方面過去 Van Dillewijn (1952), Skinner (1971) 和 Artschwager (1949) 早期已使用形態標記鑑定和區分甘蔗品種。本文檢驗 23 個甘蔗形態標記。利用 64 個甘蔗品系包括雜種，商業種和引進品種檢視形態特性的穩定性。所有品種種於東爪哇三個地點。二個地點土壤種類不同均在灌溉區，另一個在非灌溉區。結果顯示除了芽溝，其他所有的形態特性以 χ^2 值測定顯示在不同地區表現穩定，表示形態特性在不同環境下可以做為品種鑑定的可靠工具。

THE PERFORMANCE OF FAMILIES IN BREEDING FOR QUALITY TRAITS IN SUGARCANE

甘蔗品質性狀育種組合間之表現

J. A. Mariotti, M. I. Cuenya and M. B. Garcia De Salas

UIMCA (INTA-EEAOC), Tucuman, Argentina

亞熱帶地區育成早熟甘蔗是選種的一個主要目標。在這些地區採收季可由秋天一直延續到春天，因此中晚熟品種也在選種目標範圍內。通常利用糖分和蔗汁純度做為成熟指標，並且在育種上用來選拔組合和其後代的性狀。本文利用 10 個雙雜交組合來評估育種過程中與選拔甘蔗品質性狀有關

的組合表現。每個組合隨機選取 32 個後代，植於 T₃ 的重複區，評估其蔗產量指標和數個質性狀。結果顯示組合間所有質的性狀如錘度、糖度、纖維、鉀和灰分/錘度具有中高或高的遺傳力，表示組合選拔是有效可行的，但組合內的機差顯著則組合選拔無效。早熟和中晚熟品種質的性狀大部份可以重現，可見從早期到晚期選拔都有效。除了還原糖外，大部份質的性狀在組合與成熟的各階段間並無顯著交感作用，此試驗發現顯著的負相關存在於糖度 (POL) % 和還原糖之間，而顯著的正相關存在於糖度 % 和纖維之間，灰分/錘度比與纖維成負相關。

A NEW PROPOSAL FOR CLONAL EVALUATION BASED ON DIRECT ANALYSIS AND NIR SPECTROSCOPY

一個品系評估的新方法-依據 NIR 光譜圖

A. Amaya, J. Larrahondo, M. Oliveros, H. Rangel, C. Moreno, C. Viveros, C. Briceno, and W. Pena

CENICANA, Colombian Sugarcane Research Institute, Cali, Colombia

直接分析甘蔗決定甘蔗糖度%，在哥國和南非是一個很重要的選種法。以近遠紅光 (NIR) 光譜圖來代替目前糖份分析在若干國家已執行。本文以改良的直接分析法來估算糖度% 既快速，可靠且便宜。糖份是選種的一個重要標準，改良的直接分析法簡稱 CENIAD 乃根據田間甘蔗莖取下上、中、下三部位分析。CENIAD 分析結果糖度% 之間之直線相關係數達 0.98。CENIAD 每次分析各品種只須取 10 支蔗莖，從上、中、下三部份共取下直徑 2.5 公分碎片 30-33 片(約重 300 公克)，所以運回實驗室方便，分析工作快速且在實驗室可分析大量樣品。以 Infracizer 500 Bran-Luebbe 和 NIR systems 6500 分析錘度和糖份。

PRODUCTION OF ELECTRICITY FROM BAGASSE IN MAURITIUS

摩里西斯糖廠使用蔗渣發電之介紹

K.T.K.F. Kong Win Chang, L.J.C. Autrey and L. Wong Sak Hoi

Mauritius Sugar Industry Research Institute, Reduit, Mauritius

摩里西斯利用蔗渣發電的演化歷史回顧，並介紹各式政策或誘因以促使糖廠利用蔗渣發電。誘因包括補貼政策、販售蔗渣的租稅優惠、販售電力的所得稅優惠、以及發電設備投資時程本計算的優惠待遇等。在摩里西斯，糖廠發電量在過去 20 年間增加了約 120 倍，自 1981 年的 31 GWh 到 2000 年的 360 GWh，約相當於 21 萬噸進口煤。藉助世界銀行的支持，摩里西斯成立能源委員會，計畫在 2005 年將糖廠發電量提升至 2030 GWh 約相當於摩里西斯 90% 的總用電量。

A FACTILE TITRIMETRIC METHOD FOR ESTIMATION OF ACONITIC ACID IN CANE JUICE

簡易滴定法測定蔗汁中附子精酸濃度

D. Mane Jyoti, S.C. Barge and S.P. Phadnis

Vasantdada Sugar Institute, Manjari, Pune 412 307, Maharashtra, India

附子精酸在蔗汁中濃度不高，在製糖過程中大部分會存留於糖蜜中。基於糖蜜中附子精酸可能成為有價值之商品（註：附子精酸多用於塑膠與橡膠工業），因此作者提出此論文報導快速分析方法。此法與 HPLC 比較可以得到與 HPLC 分析結果成正比例之分析數據，但是並非絕對濃度，因此僅能使用於快速分析做為參考用。此外，這個分析方法需使用金屬鉛，其實用性令人懷疑。

ENVIRONMENTAL MANAGEMENT IN THE SUGAR INDUSTRY

製糖工業之環境管理

D.B. Sapkal, Deepali Nimbalkar and B.B. Gunjal

Vasantdada Sugar Institute, Manjari, Pune 412 307, Maharashtra, India

蔗糖的原料為甘蔗，製糖時的第一要務為維持環境清潔。製糖過程同時產生一些有用的副產品如蔗渣、糖蜜與濾餅等，可以為糖廠帶來可觀的收入。對糖廠而言，設置污染防治設備處理液體、固體與氣體廢棄物是必須的。此篇論文說明廢棄物種類、可能的用途等。（譯按：粗淺的描述製糖工業中所產生之固體廢棄物、液體廢棄物與廢氣之處理，毫無技術層面可言。在副產品開發論文發表會，這是一篇相當不適當的論文。）

A GAS CHROMATOGRAPHY APPROACH TO THE DETERMINATION OF FREE AMINO ACIDS IN SUGARCANE JUICE

氣相層析法分析蔗汁中游離胺機酸含量

A.V. Yewale and M.R. Shivade

Sugar Chemistry Division, Vasantdada Sugar Institute, Manjari, Pune 412 307,
Maharashtra, India

蔗汁收先去除蛋白質後，再利用陽離子交換樹脂吸附游離胺機酸，經脫附後得游離胺機酸。利用酯化使游離胺機酸成為低沸點化合物，再利用氣相層析法分析蔗汁中游離胺機酸含量。此方法以應用於生長中甘蔗的游離胺機酸測定，其應

用價值可能可以進一步提升。(譯按：此發並非首創，但是首次應用於蔗汁中。此分析方法似乎無法有效分離 Lysine 與 Arginine。)

PRODUCTION OF BIODEGRADABLE PLASTIC (PHB), SUGAR AND ETHANOL IN A SUGAR MILL

糖廠同時生產生物可分解塑膠、糖與酒精

Carlos E.V. Rossell, Roberto V. Monato, Paulo E. Mantelatto and Manoel Regis Lima Verde Leal

Centro de Tecnologia Copersucar, Cx.Postal 162 Piracicaba SP Brazil 13400-970

巴西糖廠嘗試利用糖廠生產過程中所產生之糖蜜、電力(汽電共生)同時生產酒精與生物可分解素叫 PHB (poly 3-hydroxybutyric acid)。據作者報導，一個可年產 50 公噸 PHB 之試驗工廠已經設立並運轉。PHB 之商業生產事實上已經超過 10 年，但是都處於試驗狀態，因為價格過於昂貴，無法與石油化學產品(PP 或 PE)競爭，除非政府以法規限制塑膠之來源，短時間內仍然無法與石化產品相提並論。

NATURAL GLUCANS AND MANNITOL FROM SUCROSE

以蔗糖生產葡聚糖與甘露醇

Donal F. Day, Sun Kyun Yoo and Chang-Ho Chung

Audubon Sugar Institute, Louisiana State University Agricultural Experiment Station,
Baton Rouge, La. 70703-7305, USA

利用短鏈寡聚糖圍攻功能性食品已漸漸成為一種趨勢。目前生產功能性短鏈寡聚糖的方法有兩種：一為利用酸或酵素分解多醣；另一種方法為酵素轉化反應。最有用(或最有效)的短鏈寡聚糖是聚果糖或是帶支鏈的葡聚糖。利用醱酵方法製造與生產短鏈葡聚糖。於傳統生產聚葡萄糖之醱酵培養中，添加適當之單糖分子使酵素催化之聚合反應停止，因此得到短鏈聚合物，這些短鏈葡聚糖可促進 *Bifidiobacterium* (雙歧桿菌) 生長，但不會被病原菌利用。在醱酵培養中使用蔗糖，其中蔗糖分解後所產生之果糖同時被轉化成甘露醇，可以藉低溫結晶方式分離。

**FLOCCULENT YEAST POPULATION DYNAMICS IN A CONTINUOUS
TOWER REACTOR FOR ETHANOL FUEL PRODUCTION**

利用凝集性酵母菌連續式生產酒精

**Marcelo Caldeira Viegas, Cibele Tosin Stroppa, Silvio Roberto
Andrietta, Maria Da Graca Stupiello Andrietta, and Jose Paulo
Stupiello**

FEQ/UNICAMP, Brazil

FEA/UNICAMP, Brasil

CPQBA/UNICAMP CP 6171 B. Geraldo – CEP 13083-970

STAB, Brazil

分析六種凝集性酵母菌使用於生產規模醱酵槽中、其生長動態之變化。所使用的菌株先前經觀察與試驗都發現有凝集的現象，並且其醱酵效率達到工業應用的層次。將兩個玻璃反應槽串連，供應以蔗糖為基礎的合成培養基，經 15 天培養，酵母菌株經計算其數目（使用另一種培養基），發現六種菌株都呈現不同菌落型態。其中四種酵母菌，在連續式生產後被發現喪失凝聚性，因此作者建議在實際用於生產前應該先進行培養基與培養條件探討，使凝聚性維持。

**PREPARATION OF DEAE-BAGASSE: EVALUATION OF ITS
DECOLOURISING ACTION ON SUGARCANE JUICE**

利用改質蔗渣進行蔗汁去色

J.D. Mane, D.L. Kumbhar, V.M. Bhandari and S.P. Phadnis

Vasantdada Sugar Institute, Manjari, Pune 412 307, Maharashtra, India

Chemical Engineering Division, National Chemical Laboratory, Pune 411 008, India

以化學方法利用 diethylaminiethyl chloride 進行蔗渣改質，使其帶有正電荷。此種改質蔗渣被嘗試應用於清靜後蔗汁的去色、去濁與去除糖類以外之分子。與以前使用的方法比較，此法可以更有效增進品質。利用此方法所生產的改質蔗渣比在酸性環境中改質的蔗渣帶有更多正電荷。因此，這裡所提出的方法不僅更有效的去色，而且有效的去除濁度與糖以外的雜質，此外，對蔗汁去色也有幫助。

BIOCHEMICAL DECOLOURISATION OF BIOMETHANATED DISTILLERY SPENTWASH

廢醪之生物化學去色

S. Dhamankar Vandana and U. Patil Prakash

Vasantdada Sugar Institute, Manjari, Pune 412 307, Maharashtra, India

利用黴菌的選育種 *Aspergillus niger* UM2 在厭氧環境下進行廢醪去色的研究最佳的黴菌生長與除色條件為 pH 值 4.5、溫度攝氏 30 度，並且有適量的碳源與氮源。最佳去色能力為 80%，並能去除 76.4% 的化學需養量 (COD)。所分離的黴菌被發現能有效分解約 21-22% 諸如 melanoidin, caramels 及鹼性分解產物等呈色物質。利用凝膠滲透層析分析 melanoidin, caramels 及鹼性分解產物等，發現使用的黴菌可以稍微降解前述分子，並產生較小分子量的新分子。欲達成最佳去色效果，92-98% 去色與 94-96% 去除 COD，必須讓廢醪先經過以下的化學處理：氧化鈣、氫氧化鈣、漂白粉與 0.5% 過錳酸鉀溶液。

SUGAR LOSSES CAUSED BY MICR-ORGANISMS

微生物造成之糖損失

B.S. Purchase

Sugar Milling Research Institute, University of Natal, Durban, South Africa

對整個蔗糖生產過程中可能由微生物所導致之糖損失作一回顧。微生物造成之損失包括甘蔗疾病、附著於壓榨機械之微生物生長、蔗汁中微生物族群、高溫菌與存在於糖蜜中之耐滲透壓酵母等。

ASURVEY OF POST-HARVEST BIOLOGICAL LOSSES OF SUCROSE IN INDIAN SUGAR FACTORIES: AN EMERGING CHALLENGE

印度製糖廠中微生物導致損失之調查：逐漸浮現的挑戰

P.G. Morel Du Boil

Sugar Milling Research Institute, Durban, South Africa

在印度，產糖效率降低主要是甘蔗品質差（未成熟），大量不新鮮或未成熟甘蔗（10-20%）、夾帶雜物（3-10%）以及採收後至壓榨的長停滯時間已經是產糖效率降低的主要原因。此為，不好的衛生環境加上不當使用滅菌劑都造成製糖成效的進一步降低。為了提升製糖效率，採收前甘蔗成熟度檢測、甘蔗採收時原料

品管法規修正、停閉 Cane Centre system 與使用廣效性滅菌劑等都必須正確實施。

LOSSES ASSOCIATED WITH POST-HARVEST AND PRE-DELIVERY CONDITIONS

甘蔗採收後至運輸前之損失

P.G. Morel Du Boil

Sugar Milling Research Institute, Durban, South Africa

甘蔗採收後在採收過程中所造成的傷口可能感染微生物，這些微生物約造成 1-3% 蔗糖損失（最嚴重時可以超過 10%），微生物可能將蔗糖轉化成多醣（葡聚糖）、寡糖或酒精，南非曾嘗試使用抑菌劑，但屬試驗性質，未大規模實施。

(五)心得與建議

- 1、研討會中有一新趨勢，就是由粗砂工場直接生產白糖，如此，就可減少興建煉糖工場，降低成本。但要達到此目標，高品質糖漿的獲得很重要，因此，要如何去除蔗汁中的雜質及色素，甚至在製程中，如何降低色質生成的機會等，均需新技術的支援。目前國外大都研用薄膜過濾及層析技術，但成本仍太高。建議本公司亦朝此方向研發，迎頭趕上。另外，也可由粗砂工場直接生產白糖的製程中，利用精糖漿生產高品質二砂，提供食品業者使用。
- 2、台灣糖廠都設有汽電共生系統，以燃燒蔗渣發電，提供工廠使用，但甘蔗收穫完畢後，由於沒有蔗渣作為燃料，汽電共生系統也跟著關閉，十分可惜。此次研討會中，巴西的糖廠已在研發利用夾雜物為替代性燃料，不但解決夾雜物的出路問題，亦可增加生質能源的利用。建議公司可注意此研發的最新動向，研用其成果如夾雜物的回收、包裝及運輸等技巧，來提升本公司糖廠的發電量，減少電費支出。
- 3、台灣每年生產大量的生質廢棄物如蔗渣及稻殼等，如能利用其來發電，對能源短缺的台灣，會有很大的幫助。而生質是可再生的，不像石油及煤炭，用久了會枯竭，因此，生質能源的利用是頗具潛力的事業。此次研討會中，對能源的研發亦相當的重視，其中，巴西的糖廠利用生質合成式燃氣渦輪機（BIG/GT）來發電產生電熱能，效果頗佳。建議本公司可投入此方面的研發，除了可提高糖廠的發電效率外，亦可為本省許多農業廢棄物創造出路。
- 4、蔗糖中含有多醣類如 starch、dextran 或 sarkaran 等，此種物質對製程中的過濾單元有不利的影響，而形成瓶頸，使煉糖廠的溶糖量降低，影響產能。本研討會中，南非糖研所投下巨資，進行以 HPAEC-PAD 的方法，迅速分析各種多醣類，並監控蔗汁和蔗糖中多醣類的分佈量及其在季節性和地理性上的差異。建議公司亦可建立此種分析設備及方法，以了解各進口糖中多醣類的正確含量，作為購買原料糖的重要參考資料。
- 5、台灣宿根甘蔗產量低的主要原因，就是機械化作業導至土壤壓實及蔗苗發芽不良。澳洲專家以 1.8m 植溝種二行甘蔗，發現可增產並減少土壤壓實。且依其試驗，以 1.8m 植溝種雙行蔗可行性頗高，開溝及培土作業只須將農具略為調整即可進行。建議公司可往此方向研發。
- 6、台糖亦曾以 Dalapon 及 Glyphosate 來做甘蔗成熟劑促進甘蔗成熟，效果均不錯。但以直昇機噴施仍須考慮成本，故未繼續推廣。建議公司效法研討會中的方法，考慮以灌溉水來施用甘蔗成熟劑，或許有不錯的成效。

- 7、甘蔗育種最重要目標之一是糖分和抗病性，研討會中有專家利用 Est-衍生的 RFLP 可以標記此二性狀的基因位置，將可在初期選拔即時淘汰不抗病且糖分低品系，且可將高糖基因增殖後轉殖到高產低糖抗病品系，使育種期縮短。分子標記用來輔助甘蔗育種雖然可行，但大量的材料分析費及人工損耗，將是使此方法推廣的最大阻力。建議公司可在技術方面投入部份實用性研究，待分析技術有降低費用的重大突破後，即可適時切入應用。
- 8、研討會中有專家報導，變種的黃銹病在澳洲造成極大的損失，台灣 86 年曾發生黑穗病變種，由於控制得宜，損失有限，今年更推出抗第三型黑穗病的 ROC27。而抗病育種是抵抗病害最有效的途徑，建議公司未來仍可以最少的人力及物力，維持抗病品種的誕生。
- 9、副產品僅有一篇有關蔗渣之利用，但缺糖蜜利用之研究。整體而言，研究方向趨向於生產衍生性產品，即如何提高蔗糖之附加價值。就新產品發展而言，涵蓋使用蔗糖為原料以生產寡糖、生物可分解塑膠等，此與本公司發展的方向相吻合。本次會議有數十個產糖國參加，雖然多數為熱帶開發中國家，但是仍有先進國家如美國、澳洲等地區研究人員參與，這些先進國家製糖工業所遭遇問題與臺灣較相近，會議過程中與其研究人員溝通不僅有助於瞭解其產業發展方向，並可增進友誼。

四、附錄：台糖公司參加的論文內容介紹

PROCESS MODIFICATION FOR THE PRODUCTION OF GOLDEN B-GRADE WHITE CRYSTAL SUGAR

H.K. Sheen¹, C.M. Huang¹, R.Y. Chang¹, W.C. Chen¹, L.H. Lin²,
S.Y. Hsiung², J.H. Liang²

ABSTRACT

Golden B-grade white crystal sugar, with low-sediment content, has been successfully developed by Taiwan Sugar Corporation (TSC). Most processes and equipment for the traditional production of plantation white sugar were adopted. However, several conditions such as lime requirement, optimum pH, and temperature were modified. The resulting product then met consumer requirements for low sediment and preferred color and flavor.

Keywords: Golden B-grade white crystal sugar, defecation, carbonation, plantation white sugar

INTRODUCTION

The cheapest way to produce edible B-grade white crystal sugar is by defecation (Yan, 1979a). By this process, defecated cane juice is first passed through clarifier and evaporator units. The concentrated juice is then directly pumped into the crystallization unit as the raw material for boiling. The unfiltered clarified juice contains a few nonsugars such as pith, iron rust, and colloids. Therefore, the sugar preferred quality would be negatively and greatly affected. Recently in Taiwan, customer requirements pertaining to food additives have risen greatly due to the increase in the standard of living. Hence, the carbonation method and equipment used in the traditional production of high-quality B-grade white crystal (GBWC) sugar for specific or general customers was studied. Taiwan Sugar Corporation has a long history of producing plantation white sugar by the carbonation process. The main function of this process is to remove impurities, including pigments in mixed juice by the formation of calcium carbonate. In order to increase the impurity-removal rate, a modified double-carbonation process was employed (Chen, 1985). With this method, the supply of carbon dioxide to the first defecated juice was suitably

controlled to prevent the formation of calcium bicarbonate. As a result, the impurities absorbed by calcium carbonate would not be released again due to the formation of calcium bicarbonate, which has higher solubility than calcium carbonate (Yan, 1979b). Generally, the reducing sugars in the first-saturated carbonated juice can easily produce colour through high temperature. Therefore, temperature is a very important factor to be controlled in the production of plantation white sugar. However, in the manufacturing process for edible B-grade white crystal, it is necessary to retain the original color and flavor of cane juice, and this is different from that of plantation white sugar that used the carbonation process. Hence, in this study some modifications in the carbonation process for the production of golden B-grade white crystal were necessary.

MATERIALS AND METHODS

Temperature and pH effects on filtration of first-carbonated juice in simulated CO₂ saturation tank

A 500-ml sample of mixed juice, which was heated, limed and partially carbonated in the sugar mill, was put into a simulated CO₂ saturation tank to increase its carbonation. The samples of the mixed juice were heated to 60, 65, 70, 75, 80 °C, respectively, and their pHs were adjusted to 9.6, 9.8, 10.0, 10.2, 10.4 and 10.6, respectively, by carbon dioxide. A 300-ml sample of carbonated juice was filtered. The filtration rate was recorded and the pH, Brix, polarization, color, calcium oxide and invert sugar in the filtrate were analyzed.

Mill production of golden B-grade white crystal sugar

1. Modified operation conditions

- (1) Changed temperature of mixed juice
- (2) Changed pH of the first and second carbonated juice
- (3) Changed pH of the sulphitated syrup in the diluted juice sulphitation tank
- (4) Carbon dioxide in place of sulfur dioxide was used in the diluted juice sulphitation tank
- (5) Changed boiling system

2. Modified equipment of carbonation

- (1) In order to reduce lime consumption and to maintain juice agitation in the carbonation tank simultaneously, one valve had to be added ahead of the carbon dioxide pump inlet to allow for introducing air in place of some of the carbon dioxide.
- (2) Raising the juice overflow height increased the retention time in diluted juice sulphitation tank in order to optimize the reaction carbonation rate (Chen, et al., 1980).

3. Process establishment of edible B-grade white crystal sugar

The process for production of edible B-grade white crystal sugar is similar to that of production of plantation white sugar.

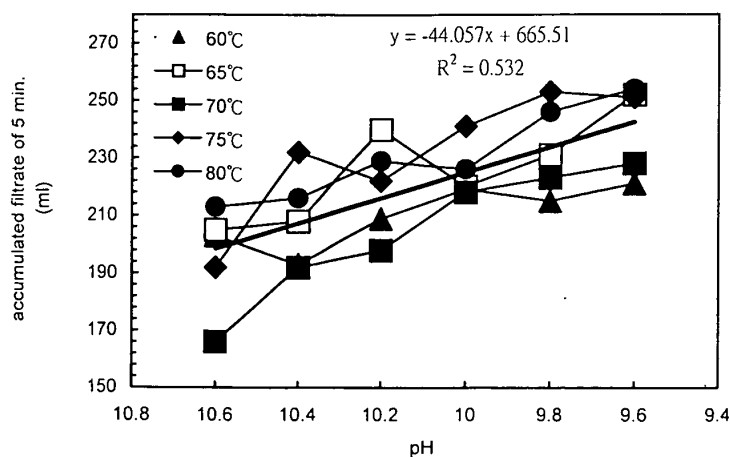


Figure 1. The effect of temperature and pH on the filtration of first carbonated juice

RESULTS AND DISCUSSION

Temperature and pH effects on filtration of first-carbonated juice

Temperature and pH effects on filtration of first-carbonated juice are shown in Figure 1. At the same pH and using the same sample, the filtration rate at 65°C was greater than that at 60°C. Similarly, the filtration rate at 75°C was greater than that at 70°C. Obviously, increasing temperature of the first carbonated juice enhanced the filtration rate at 60-80°C. At any given temperature, the filtration rate of first-carbonated juice did not show an obvious relation to pH, but when the results at all temperature are considered together, the filtration rate increases as the pH decreases. Perhaps, it was helpful to the filtration operation when more precipitate was formed because more carbon dioxide was combined in the juice in the first-carbonation tank.

In the pH 9.6 to 10.6, the color of filtered juice rose gradually with declining pH. Color and impurities which had been absorbed by calcium carbonate in the juice were re-released because of excess carbonation. The ratio of calcium oxide on brix (CaO/Bx) decreased as pH decreased in the filtered juice. The concentration of carbonate ions would increase and pH in juice would decrease, because more carbon dioxide entered in the juice. Increasing concentration of carbonate ions in the juice may also react with residual calcium ions to form precipitate before coming to equilibrium in the juice. The ratio of calcium oxide on brix decreased when the

precipitate was filtered from carbonated juice. Calcium ion content in filtered juice could seriously affect evaporation because too much calcium ion could form scales which reduce the efficiency of heat transfer in the evaporator and thereby increase sediment in product.

Edible B-grade white crystal sugar manufactured by the sugar mill

TSC planned to manufacture golden B-grade white crystal sugar at Huwei, Nanching, and Chishan sugar mills that previously made plantation white sugar in the crop year of 98/99. This required modification of some equipment and controls used in this carbonation process and boiling system. Some difficulties have been overcome such as distribution of carbon dioxide, filtration of carbonated juice, viscosity of boiling, and adjusting condition to smoothly make the product at those mills. There were different control processes at each mill that were affected by milling capacity and original equipment, but the overall process was in accord with this carbonation process. The product met manufacturing standard of edible B-grade white crystal sugar and consumer demand for quality, including low sediment (Table 1). Obviously, these sugar mills, through modifying their processes, could also produce large amounts of golden B-grade white crystal sugar with low sediment.

Table 1. The characteristics of Golden B-Grade White Crystal Sugar

Mill	Yield (ton)	Pol. (%)	Water content (%)	Ash (%)	Color value (RBU)	Size (%)	Sediment
Chishan	4736	98.76	0.122	0.144	797	93.21	B
Nanching	3547	98.78	0.167	0.173	654	88.64	C
Huwei	2995	98.67	0.153	0.162	1008	89.01	B
Average	3760	98.74	0.147	0.160	820	90.27	B

CONCLUSIONS

The filtration rate obviously increased when the filtration temperature rose at the same ratio of juice lime. This had a beneficial effect on the carbonation and filtration process when making golden B-grade white crystal sugar. The low-sediment manufacturing process of edible golden B-grade white crystal sugar, characterized by controlling carbonation equipment, was revised so as to enable it to match the quality of plantation white sugar made from the traditional carbonation process, and thereby retaining preferred color and taste. This process will become a major manufacturing method for edible B-grade white crystal sugar.

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AN AUTOMATIC CONTROL SYSTEM FOR RECOVERING EXHAUST STEAM

L. H. Huang

ABSTRACT

Because fuel oil amounts to approximately 40 % of the cost of sugar refining, cost-effective management requires reducing fuel oil consumption. The objective of this study is to develop an automatic control system to recover the exhaust steam which builds up primarily during the sugar- refining process, due partially to the decreasing efficiency of the turbogenerator, and then to direct it to the boiler feedwater tank. The system is designed to detect the upstream pressure of the control valve and to sense the temperature of the boiler feedwater. A proportional integral derivative (PID) pressure controller and a proportional temperature controller are used in the system. Results showed that the automatic control system was stable and that the system is capable of increasing the temperature of the boiler feedwater from 50°C to 92°C, thereby saving 4.2 kl of fuel oil per day.

Keywords: boiler feedwater temperature, fuel oil consumption, exhaust steam recovery, energy efficiency, automatic control.

INTRODUCTION

The blow-off exhaust steam from sugar refining process of the Peikang sugar refinery is 3.33t/h. It is equal to a heat discharge of 488,972 kJ into the air per ton of superior white crystal sugar, thereby wasting energy and producing excessive noise, especially at night. Therefore an automatic control system was designed to recover the exhaust steam and to direct it to the boiler feedwater tank to increase feedwater temperature, thus reducing fuel oil consumption.

In this automatic control system, the upstream steam pressure detected by a pressure sensor is compared with the reference variable (command). Any difference between the two produces an error signal (actuating signal) which is used to drive the positioner and the actuator of the control valve (Liptak, 1993). Solberg et al (1965) states that the direct-contact feedwater heater (open heater) has the capacity to completely convert steam to water and to remove noncondensable corrosive gases from the feedwater. Generally, due to the stress limitations of the heater shell, steam pressure is limited to 0.5 barg. Spirax Sarco Ltd. (Anon, 1998) explain that the PID controller functions remove offset and adjust the controller response to allow rapid return to steady state. This research mostly uses automatic process control and thermal engineering technologies. Once the system is in use in the Peikang sugar refinery, it will reduce annual fuel oil costs

MATERIALS AND METHODS

1. System design and materials used

The automatic control system is designed to steadily recover the exhaust steam accumulated primarily from the sugar refining process, due in part to the decreasing efficiency of the turbogenerator. The functional components of the control system consist of a diaphragm control valve, pressure transmitter, temperature probe, PID pressure controller, proportional temperature controller, stopvalve, flowmeter, steam trapping station, vacuum breaker and direct-contact heater. The block diagram of the system is shown in figure 1. The purpose of the upstream-sensing device which monitors control valve pressure is to maintain exhaust steam pressure and flowrate in order to meet the requirements of the sugar refining process. The feedback temperature control limits the temperature of the boiler feedwater. In other words, the amount of the exhaust steam recovered to the feedwater tank is determined by

controlling the upstream pressure of the control valve and the temperature in the boiler feedwater.

The exhaust steam produced by the turbogenerator is used in the sugar refining process which requires an average flowrate of 22t/h with a pressure of 0.3 barg. The automatic discharge valves mounted on the turbogenerator begins to blow off at a back pressure of 1.0 barg. A 200 mm (8 inch) diameter pipe, used by the automatic control system, is connected to a 500 mm (20 inch) diameter pipe through which the exhaust steam flows to the manufacturing process. A stopvalve mounted at the fore-end of the control system is fully opened when the system is in operation. Two distributors are located within the circular 90 ton feedwater tank to evenly distribute the exhaust steam in the tank, assuring rapid condensation of steam and efficient water heating. The feedwater tank and all the pipes of the system are well insulated.

A pressure transmitter is mounted at the inlet of the control system to detect the exhaust steam pressure injected in the sugar- refining process and transmits 4-20 mA current to a PID pressure controller. Then the signal is compared with the command set on a PID pressure controller. An autotune device adjusts the PID parameters to the optimum values to protect from controller instability and overshoot. The PID pressure controller is used to compensate for load changes and maintain a zero offset under steady-state conditions. When the actual signal exceeds the command, an error (actuating) signal results, which drives a valve positioner and actuator, thus lifting the control valve seat and letting through the exhaust steam.

The temperature probe is positioned at the outlet of the boiler feedwater tank. Before the feedwater temperature reaches the set temperature on the temperature controller, the PID pressure controller controls the release of steam. Once the feedwater temperature reaches the set temperature, the temperature controller overrides the pressure controller and stops the release of steam to the feedwater tank. If the compressed air supply abruptly stops or the PID pressure controller malfunctions, the control valve will be closed immediately.

2. Experimental methods

The spindle lift percentage of the control valve can be adjusted and set on the PID pressure controller in advance. An exhaust steam set point pressure of 0.6 barg was set on the PID pressure controller. The pressure can be adjusted a little higher or lower but not less than 0.4 barg. One pressure gauge is mounted in front of the

stopvalve to indicate the exhaust steam pressure occurring in the sugar refining Process. A second pressure gauge indicates the steam pressure downstream of the control valve. A temperature of 92°C is set as command on feedwater temperature controller, which can be adjusted up to 96°C for this study. Fuel oil consumption needs to be recorded to ascertain the automatic control system economical benefits when it is in operation.

RESULTS AND DISCUSSION

Results showed that 80 tons of exhaust steam were recovered to the feedwater tank per day. The temperature of the exhaust steam directed to the feedwater tank was 105°C, and its enthalpy was 2,684 kJ/kg. In other words, the averaged flowrate of the exhaust steam through the control valve was 3.33 t/h, thus increasing the feedwater temperature from 50°C to 92°C. The percentage of the spindle lift of the actuator was set 60% of its maximum value. The spindle stroke is 5 cm. During test, the spindle lift was 3 cm under steady state. The spindle movement was stable by observation. The pressure indicated by the downstream pressure gauge was 0.2 bar which had no effect on the 9-mm feedwater tank wall because it was only slight pressure. The distributor eliminated water hammer and operated with minimum noise.

When the feedwater temperature reached 92°C, the control valve automatically closed due to the set value on the proportional temperature controller. At this time no exhaust steam could flow through. The control valve opened again when the feedwater temperature was lower than the set value.

Results showed that this automatic control system proved stable with fast response. Results also showed that after the control system was in operation, 4.2 kl of fuel oil per day were saved. Hopefully, it will save fuel cost of U.S.\$236,250 every year, thus lowering the cost of sugar refining.

The recovered exhaust steam from the manufacturing process was attributed primarily to cuts, massecuite discharge, and products purging; and partly to the decreasing efficiency of the turbogenerator, which was consuming 0.96 kg of live steam per kWh, more than the normally required and equivalent to an additional 20 tons per day of steam.

CONCLUSIONS

An automatic control system has been implemented which controlled exhaust steam supply pressure by diverting excess exhaust steam to heat the boiler feedwater. As a result, about 80 tons of exhaust steam per day were automatically recovered to the feedwater tank, thereby increasing the feedwater temperature from 50°C to 92°C, saving 4.2 kl of fuel oil per day. The automatic control system will reduce fuel oil cost by U.S.\$236,250 every year, thus lowering the cost of sugar refining.

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PRDUCTION OF FRUCTOOLIGOSACCHARIDES FROM SUCROSE SYRUP BY A MICROORGANISM TSC-FOS1

Huang, i-j., chen, p.-l., chang, c.-y., wang, G.-S., AND Wang, J.-S.

ABSTRACT

Fructosyltransferase activity from a microorganism (designated TSC-FOS1) capable of converting sucrose to fructooligosaccharide (FOS) was investigated. Using cultured cells, we were able to produce FOS from 50% sucrose syrup. Maximum cellular activity of FOS production was determined to be pH 6-7 and 65 °C. Enzyme activity was stable within a very narrow pH range, pH 5; and it was sensitive to higher temperature. Optimum reaction condition for FOS production was determined to be at pH 5.5 and 55 °C, after considering stability factors. The activity was inhibited by FeCl₃, Al₂O₃, and methanol. The FOS content produced by the cultured cells was between 55-60% (FOS to total carbohydrates).

Keywords: FOS, fructosyltransferase, sucrose

INTRODUCTION

Fructooligosaccharides (FOS) are considered a “health food” because of their special properties which lower serum lipids, increase intestinal calcium absorption, and indigestible ingredients (calorie-free and safe for diabetics) (Hidaka *et al.*, 1986; Stamp, 1990; Tokunaga, 1989). They benefit the host by stimulating the growth of beneficial microflora (e.g. acidophilus and bifidum). FOS are composed mainly of 1-kestose (GF₂), nystose (GF₃), and fructofuranosyl nystose (GF₄), in which fructosyl units (F) are bound at sucrose (GF). These bonds cannot be broken down by enzymes in the human digestive system. Because the sweet taste is similar to table sugar, they are alternative sweeteners for consumers desiring healthier and calorie-controlled foods.

FOS is produced by the conversion of sucrose through fructosyltransferase (EC 2.4.1.9). They are found in many higher plants, such as sugar beet, onion, and asparagus (Allen and Bacon, 1956; Henry and Darbyshire, 1980; Shiomi *et al.*, 1976; 1979). Industrial-scale FOS production is done mainly with fungal enzymes from either *Aureobasidium* sp. (Yun *et al.*, 1992; 1990) or *Aspergillus niger* (Hidaka *et al.*, 1987; Hidaka *et al.*, 1988). Meiji Seika Co. (Japan) was the first to commercially produce FOS (Neosugar) by *Aspergillus niger* enzyme.

In this study, we characterized the fructosyltransferase activity of an isolated microorganism that is capable of converting sucrose into FOS. This enzymatic system is used to manufacture FOS in commercial scale by the Taiwan Sugar Corporation.

MATERIALS AND METHODS

Cultivation conditions. A fungus (TSC-FOS1) from our microorganism banks had been identified for its ability to produce FOS. This microorganism was inoculated into a 1-L shake flask from a slant, and grown for two days at 27 °C. The culture was subsequently inoculated into 5-L and 100-L fermenters containing selected media. At the end of fermentation, the broth was centrifuged to harvest cells. The moisture content of the collected cells was 70-72%. The cells were dried at 35 °C and ground into powder for enzyme assay.

Enzyme assay. To determine the enzyme activity, harvested cells were added to a 50 % (w/v) sucrose solution at various pHs and temperatures. At the end of the reaction, the enzyme was inactivated in boiled water for 10 min. The end products were diluted 20-fold with deionized water and filtered before being subjected to high performance liquid chromatography (HPLC) to determine the amount of each carbohydrate. The activity was defined as the ratio of FOS, which were produced in

the reaction, to the total carbohydrates in solution. For pH-effect study on the enzyme, Malvaine buffer and Clark and Lubs buffer were used for the differing pH ranges.

High performance liquid chromatography (HPLC). HPLC analysis was performed with a Sugar PAK-I column (Waters, USA). The injected samples were eluted with 500 mg/L Ca-EDTA (Sigma) at 0.6 ml/min and detected with a refractive index detector (RI). The column temperature was kept at 90 °C during separation. Total FOS produced by the cells was calculated based on the deduction of the residual sucrose, glucose, and fructose at the end of the reaction from the initial sucrose.

RESULTS

Maximal FOS production activity. In order to obtain maximum FOS production from this microorganism, we sought to determine the optimum reaction conditions. Because the majority of the fructosyltransferase existed in the cells (data not shown), only the intracellular activity was investigated.

Cells harvested from the fermentation process were dried at low temperature and kept at room temperature until use. The effects of pH and temperature on enzymatic activity are shown in Fig. 1. Temperature from 30 to 70 °C were tested for FOS production for one hour as described in Materials and Methods. As shown in Fig. 1A, maximum activity was found at 65 °C. The effects of pH on the enzyme were performed at pHs between 3 and 9 for one hour. As shown in Fig. 1B, pH 6 to 7 resulted in maximum activity. The activity rapidly dropped off at pH above 8.

Stability test. The effect of temperature on the stability of the enzyme was studied by incubating the cells in pH 5.5 at various temperature from 40 to 70 °C for one hour, then subjected to FOS-production assay for one hour. The activity rapidly decreased as the cells incubated at higher temperature (Fig. 2A). This indicated that the enzyme is temperature-sensitive. After incubation at 55 °C for one hour, 22 % (w/w) FOS was synthesized, and that was only 13 % at 60 to 65 °C. Therefore, 55 °C was chosen for further studies, although the maximum activity was found at 65 °C. The cells were also incubated at 55 °C for one hour with various pH (3 to 9) conditions, then subjected to the reaction described above for stability studies. As shown in Fig. 2B, the enzyme is most stable within a narrow pH range-around 5.

Effect of chemicals. Investigation of reaction mixtures containing various chemicals at pH 5.5 and 55 °C was conducted for one hour to study their effects on the FOS-production ability of the cells. As shown in Table 1, the activity was greatly inhibited by FeCl₃, Al₂O₃, and methanol.

Table 1. Effect of chemicals on FOS-production activity

Compound	Concentration	Relative activity (%)
Methanol	0.40 %	69.55
Al ₂ O ₃	0.1 mM	52.02
CaSO ₄ • 2H ₂ O	0.1 mM	82.97
FeCl ₃ • 6H ₂ O	0.1 mM	31.87
MgSO ₄ • 7H ₂ O	0.1 mM	96.88
Control	-	100.00

FOS production. The reaction profile of FOS production by the cells is shown in Fig. 3. During the reaction, sucrose was hydrolyzed to glucose and fructose. The amount of glucose started to accumulate, and fructose was converted into FOS as through time. The FOS content exceeded 50 % (w/w) to total carbohydrates within 3 hours. The rate of FOS production decreased and gradually stopped after glucose reached about 11% (w/v) in the reaction mixture.

DISCUSSION

Fructooligosaccharides (FOS) are indigestible, naturally occurring carbohydrates. They have drawn great attention from many laboratories because of their positive functional properties to humans and animals. Japan has the largest market; its market volume was 4,000 metric tons in 1990. This market is expected to increase. Therefore, many companies are expanding their FOS market to the United States and Europe. For these reasons, Taiwan Sugar Corporation is also interested in FOS production.

In this study we investigated the FOS-producing activity of a fungus (designated TSC-FOS1) obtained from our microorganism bank. The temperature for maximum FOS-production by the cells was determined to be 65 °C; however, its residual activity was only 38% of the control after treating cells at this temperature for one hour. As shown in Fig. 2A, the FOS-producing activity greatly decreased as the incubation temperature rose.

Although the activity at 55 °C was only 85% of that at 65 °C, the enzyme is more stable at this temperature. Its optimum reaction temperature for FOS production were determined to be 55 °C. The maximum reaction pH was 6 (Fig. 1B); however, the enzyme was most stable at pH 5 (Fig. 2B). The enzyme was stable only within a narrow pH range. The FOS-producing activity at pH 6 was only 60% of that at pH 5. The optimum pH for producing FOS was then determined to be 5.5 after both activity and stability were included in the evaluation. The activity was inhibited by some

chemicals (Table 1), including methanol, FeCl_3 , and Al_2O_3 . Similar observation was found in *Arthrobacter globiformis*. The fructosyltransferase activity purified from *Arthrobacter globiformis* was inhibited 82% by ferric ion at 1 mM of concentration (Seki *et al.*, 1989). This result is very different from that of *Aspergillus niger* (Masao *et al.*, 1989). The FOS-producing enzyme was not inhibited by ferric chloride at 1 mM of concentration.

It is known that fructosyltransferase has both hydrolyzing and transfructosylating activities. During the reaction sucrose is quickly hydrolyzed into glucose and fructose. The fructosyl residues are then transferred to sucrose to produce FOS, and glucose accumulate. Since glucose is a feedback inhibitor of fructosyltransferase (Jung *et al.*, 1989), the FOS in the syrup can only reach 55-60% of the total carbohydrates. Similar results were found in our FOS-production system (Fig. 3). After three hours reaction, the rate of FOS synthesis decreased, and the fructose gradually accumulated.

The sucrose-hydrolyzing activity was also inhibited. This system, therefore, is limited in its capacity to produce a product with high FOS content. Because of this, a two-enzyme system that included glucose oxidase in the reaction solution to raise the FOS content to 90-98% was developed (Jung *et al.*, 1993; Yun *et al.*, 1994). By incorporating glucose oxidase in the reaction solution, glucose was oxidized to gluconic acid, and the feedback inhibitor was eliminated.

The other means of producing high-content FOS syrup used column chromatography. We developed a system using 10 in-series columns of ion-exchange chromatography. With this system we obtained FOS syrup up to 90% from 60% sucrose syrup. Commercial-scale application is under development.

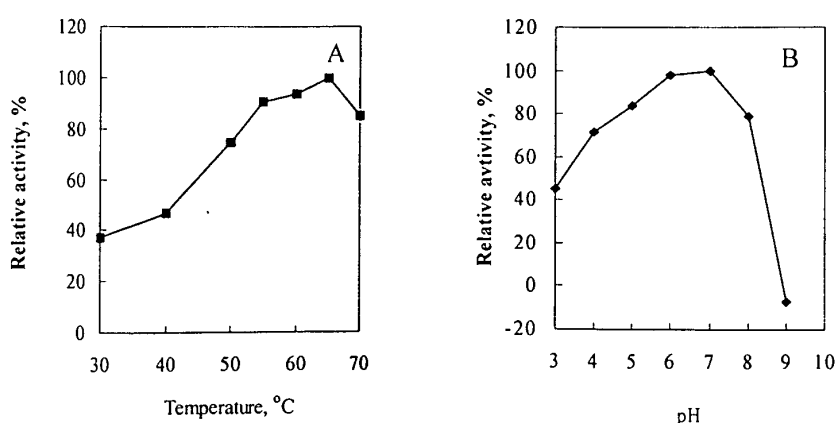


Figure 1. Effects of temperature and pH on the FOS-producing activity of the cultured cells. A: temperature effect; B: pH effect. The reactions were carried out

in a reactor containing harvested cells and 50 % sucrose with various pHs or temperatures for one hour. The amount of each carbohydrate was determined by HPLC. Activity was defined as the ratio of FOS produced to the total carbohydrates at the end of the reaction.

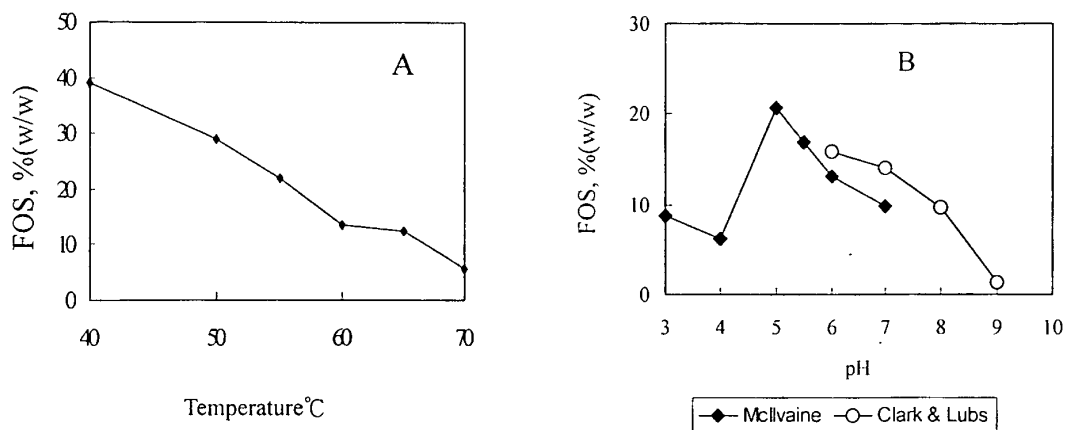


Figure 2. Effects of temperature and pH on the stability of FOS-producing activity of the cultured cells. A: temperature stability; B: pH stability. The buffers used for different pH range were as following: pH 3 to 7, 0.1 M McIlvaine buffer; pH 6 to 9, Clark & Lubs buffers. After dried cells (0.05 % [w/v]) were incubated at various temperatures or pH buffers for one hour, the enzyme solutions were added to 50 % sucrose syrup for FOS production assay for one hour. The activities were assayed as described in Materials and Methods. Activity was defined as the ratio of FOS produced to the total carbohydrates at the end of the reaction.

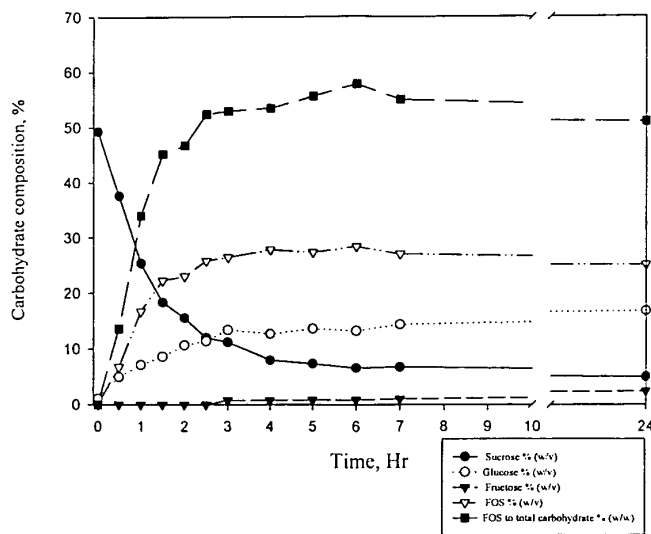


Figure 3. Reaction profile of FOS production by cultured cells. The reaction was carried out with 0.5 % (w/v) of harvested wet cells at pH 5.5, 55 °C, and 150 rpm agitation in a 5-L reactor containing 50 % (w/v) sucrose syrup. At each time point, samples were boiled for 10 min. to stop the reaction, then subjected to HPLC analysis to quantify each individual carbohydrate.

CONCLUSIONS

We reported the properties of fructosyltransferase from a microorganism. The optimum condition for producing FOS by the cells was determined to be at pH 5.5 and 55 °C. The enzyme was sensitive to ferric chloride and aluminum oxide at 0.1 mM as well as methanol at 0.4 %. We currently use it to produce FOS on a commercial scale with 10-ton reactors at Pu-Li Food Division, Product Development Department, Pu-Li, Taiwan.

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PRODUCTION OF SELENIUM YEAST

Jia-Jer Liou, Yi-Hung Lin, Wen-Ling Cheng, John Po-Wen Yang, Long-Huei Wang
Taiwan Sugar Research Institute, 54 Sheng-Chan Road, Tainan 701, Taiwan

ABSTRACT

A process for the production of high organo-selenium content yeast has been developed at the Taiwan Sugar Research Institute (TSRI). Selenium tolerant strains of *Saccharomyces cerevisiae* were obtained through traditional strain selection methods from various sources. Among the selected strains, the *S. cerevisiae* TSL-1 was found to give the best yield of organo-selenium when sucrose was used as the main carbon source. The organo-selenium content of *S. cerevisiae* TSL-1 reached about 1800 ppm after 50 hours of cultivation. A mass production process including the separation of inorganic selenium from the final product was designed and tested at a fermentation pilot plant at the TSRI.

Key Words: organo-selenium, *Saccharomyces*

INTRODUCTION

Selenium, a trace element commonly exists in soil in low level, is essential for human body to sustain normal metabolism (Schwarz and Foltz, 1957). The discovery of its role in glutathione peroxidase (Rotruck et al., 1973) triggered the interest of many researchers on the defensive function that selenium gives against oxidative stress (Combs, 1988). Deficiency in selenium causes health problems such as metabolism disorder or, in a serious condition, Keshan disease, which is a

cardiomyopathy seen in some areas where selenium is very rare in soil (Xia et al, 1994; Winnefeld, 1993; Cohen and Avissar, 1993).

Selenium has been reported as a key component of some antioxidants that can prolong life expectancy of animals and possibly human life as well (see monographs in references Burk, 1994, and Prasad, 1993). Furthermore, selenium was reported to play putative roles in preventing chronic diseases, most notably carcinogenesis by some researchers (Burk, 1994; Burk, 1989; Combs, 1988). Following the recognition of the potential functions of selenium to human health, studies on the dietary intakes of selenium and related concerns, such as using selenium as a food supplement, have also prospered (Combs, 1988).

Various studies have revealed that organoselenium, mainly selenocystein and selenomethionine in proteins, gives better bioavailability of selenium in animal tests (van Ryssen et al., 1989). More importantly, it has been reported that organoselenium is safer than inorganic selenium sources as a daily nutritional supplement (Mao et al., 1991; Haas and Velten, 1993). Thus, organic sources of selenium are superior to inorganic selenium. Some yeast strains can accumulate a significant amount of organoselenium quickly when cultivated in an environment enriched with inorganic selenium (Korhola et al., 1986; Huang et al., 1988; Xie et al., 1990). Comparing to selenium eggs or selenium vegetables, selenium yeast has obvious advantages in terms of the versatility of product forms. In this paper, the search of a yeast strain with high yields of organoselenium and the cultivation of selenium yeast will be discussed. A production process that can produce around 60 kilograms of selenium yeast per batch was developed using the selected strain.

MATERIALS AND METHODS

I. Strain

Yeast strains in the Taiwan Sugar Institute collection were used for this study, including four strains of *Saccharomyces cerevisiae*, strain W-3, H-1, TSL-1, and TSL-2, and one *Candida utilis*, strain Y-900. A commercially available baker's yeast strain acquired from a local supermarket was also used. Strains were kept in YPD agar slants that contain 2% yeast extract (Difco, Detroit, MI, USA), 1% Bacto peptone (Difco), 2% dextrose (Ishizu Seiyaku, Osaka, Japan), and 5% Bacto agar (Difco). Colonies were taken from 3-day-old YPD plate for further experiments.

II. Determination of Selenium Tolerance

Selenium tolerance of each strain was determined using sodium selenite (Merck, Germany) enriched YPD plates. A colony was taken from a 3 day old YPD plate and suspended in sterilized distilled water. Proper dilutions made out of the yeast-in-water suspension were then spreaded aseptically onto selenium enriched

YPD plates and also onto selenium free YPD plates for comparison. The number of colony on plates was then counted after two days of incubation at 30 °C to determine the colony-forming unit per ml of yeast in water suspension.

III. Cultivation Method and Cell Mass Determination

A yeast colony was taken from a 3-day-old YPD plate and aseptically transferred into a 500 mL flask containing YPD medium or YPD medium enriched with sodium selenite. Inoculated flasks were then incubated in an orbital shaker (model 706R, Hotech Instrument, Taiwan) at 30°C at 120 RPM. Cell mass was determined using dry cell weight, which was determined by taking 10 ml of suspension from the flask; after washed once with distilled water, cells were dried in a 105 °C oven, and then weighed.

IV. Quantification of Organoselenium

Dried yeast samples were washed with 0.1N hydrochloric acid or distilled water for 10 minutes and then determined their selenium contents following the fluorometric method of Alfthan (Alfthan, 1984).

V. Pilot Scale Production

Pilot scale production was performed at the TSRI using a 6000 L fermentor with 50% working volume. Cane sugar and urea were used as the main carbon and nitrogen sources. Produced selenium yeast was washed twice with water to reduce the concentration of selenium dissolved in liquid, presumably inorganic selenium, to be less than 1 ppm. Selenium yeast was then recovered using a spray drier that was custom made for the TSRI.

RESULTS AND DISCUSSION

In some preliminary studies on our yeast strains, we found out that *C. utilis* was much less tolerant to selenite than all *Saccharomyces* strains. Among the *Saccharomyces* strains, the tolerance to selenite varied. Strain TSL-2 and the baker's yeast purchased from supermarket were obviously less tolerant than others. Further investigation on the tolerance to inorganic selenium was carried out on strains W-3, H-1, and TSL-1 for up to 100 ppm of selenium. Inorganic selenium in selenite form clearly inhibited the growth of strains W-3 and H-1. Approximately 25% of strain W-3 and 80% strain H-1 cells failed to form colony on plates containing 25 ppm selenium after two days. Furthermore, the size of single colonies on plates enriched with selenium appeared to be smaller than that without selenium. The higher selenium content was on a plate, the smaller the size of colonies was. Among the tested strains TSL-1 showed the best selenium resistance. For selenium concentration less than 50 ppm, selenite did not affect the number of colony forming on plates. Interestingly, the size of single colonies of TSL-1 also decreased as the

selenium concentration of a plate increased. This indicated that selenite also inhibited the growth of TSL-1, however, not as significant as it did to strains W-3 or H-1.

It has been reported that when growing in a selenium enriched environment, a yeast cell incorporates selenium predominately into selenocystein and selenomethionine (Haas and Velten, 1993). However, it is difficult to distinguish chemically bound selenium from inorganic selenium absorbed to organic compounds (Abrams et al., 1990). The common practice to separate organoselenium from inorganic selenium is to use extractability of selenium into water or dilute acid (Arthur and Bechett, 1994; Korhola and Edelman, 1986). Although washing with water or dilute acid does not guarantee to give samples free of inorganic selenium that is physically bound to organic molecules. It is believed that inorganic selenium bound to organic molecules would be rare after washing. In the following discussion, the term “putative organoselenium” stands for the selenium content determined using the described methods, which is likely to contain both organoselenium and trace of inorganic selenium bound to organic molecules.

Batch cultures of strain TSL-1 were kept in shaken flasks to investigate the effect of inorganic selenium to cell growth based on the dry cell weight. The doubling times were 1.74, 1.86, 2.01, and 2.14 hours in cultures with initial concentrations of inorganic selenium 0, 5, 10, and 20 ppm, respectively. Cell growth in all cultures significantly slowed down 12 hours after inoculation. After 24 hours of cultivation, the cell concentration in individual cultures was 4.48, 4.16, 4.06, and 3.12 g/L corresponding to the order of increasing initial selenium concentration in the cultures. The growth rate during exponential phase and the yield of biomass at 24 hour both show that selenite ion inhibited the growth of strain TSL-1. With 20 ppm of selenium in the form of selenite, the yield of biomass at 24 hour decreased about 30% comparing to that of selenium free medium.

The time course of putative organoselenium content in strain TSL-1 cells cultivated in medium with various initial concentrations of inorganic selenium showed a common trend shared by the cultures enriched with selenium: the putative organoselenium content increased with time. The increase in putative organoselenium content was most profound between 8 to 12 hours after inoculation, which corresponded to the stage when cells enter into stationary phase from exponential phase. During stationary phase the increase in putative organoselenium was insignificant. Since selenium is mainly incorporated into selenocystein and selenomethionine, it is expected that putative organoselenium content could increase only when cells are actively producing amino acids. The final putative organoselenium content, which was 190, 580, and 1190 ppm for cultures enriched

with 5, 10, and 20 ppm inorganic selenium, respectively, increased with the quantity of inorganic selenium initially added into the cultures.

It is not clear if yeast cells can distinguish selenium from sulfur. However, selenium seems to compete with sulfur for the positions in cysteine or methionine (Haas and Velten, 1993). The result of our experiments showed that within 20 ppm of inorganic selenium added, the rate of incorporation of selenium into organic molecules in yeast cells was roughly proportional to the concentration of selenium in the medium. It is likely that the concentration of inorganic selenium was significantly lower than sulfur containing molecules or ions in the culture medium. If the initial concentration of selenium was further increased to some certain extent, the final putative organoselenium content might increase proportionally to a level higher than 1200 ppm. However, at further raised inorganic selenium level, the growth of yeast cells would significantly slow down. Although a medium containing high initial concentration of inorganic selenium may yield a putative organoselenium content of more than 1200 ppm, no medium higher than 20 ppm was tested for the consideration of downstream processing.

The pilot trials of production were performed using a 6000 L fermentor with 50% working volume. Carbon source was fed with a scheduled rate that was intended to keep the yeast cells growing. Sodium selenite solution was added in after the concentration of yeast reached about 5-7% to avoid the retardation of growth at the early stage. After about 40 hours, the biomass reached the maximum of around 20 grams per liter and started to slightly decrease. The organic selenium content of harvested yeast was in the range of 1700 to 1800 ppm on a dry weight base.

CONCLUSION

A strain of *S. cerevisiae* was screened and isolated from the yeast collection of the TSRI, which is able to accumulate 1150 µg putative organoselenium per gram dry cells within 24 hours. The rate of accumulation of putative organoselenium in yeast cells depends on the initial concentration of inorganic selenium added to culture media. Within 20 ppm of inorganic selenium added into culture media, the final content of putative organoselenium in selenium yeast increases with the initial level of inorganic selenium. The pilot trials using the selected strain yielded 20g/L selenium yeast that contained up to 1800 ppm organic selenium.

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