行政院國家科學委員會補助
大專學生參與專題研究計畫研究成果報告

計畫名稱：球將乾燥粉末對於 Hedgehog 訊息活化誘導小鼠前列腺癌之嘗試性治療研究

執行計畫學生：陳胤廷
學生計畫編號：NSC 98-2815-C-040-025-B
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指 導 教 授 ：張菡馨

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大專生專題研究計畫成果報告

球薑乾燥粉末對於 Hedgehog 訊息活化誘導小鼠前列腺癌之嘗試性治療研究

計畫編號：98-2815-C-040-025-B

學生： Yin-Tin Chen (陳胤廷)

計畫指導教授：Han-Hsin Chang (張菡馨) 副教授

執行期間：2009 年 7 月 1 日至 2010 年 2 月 28 日

一、中文摘要：

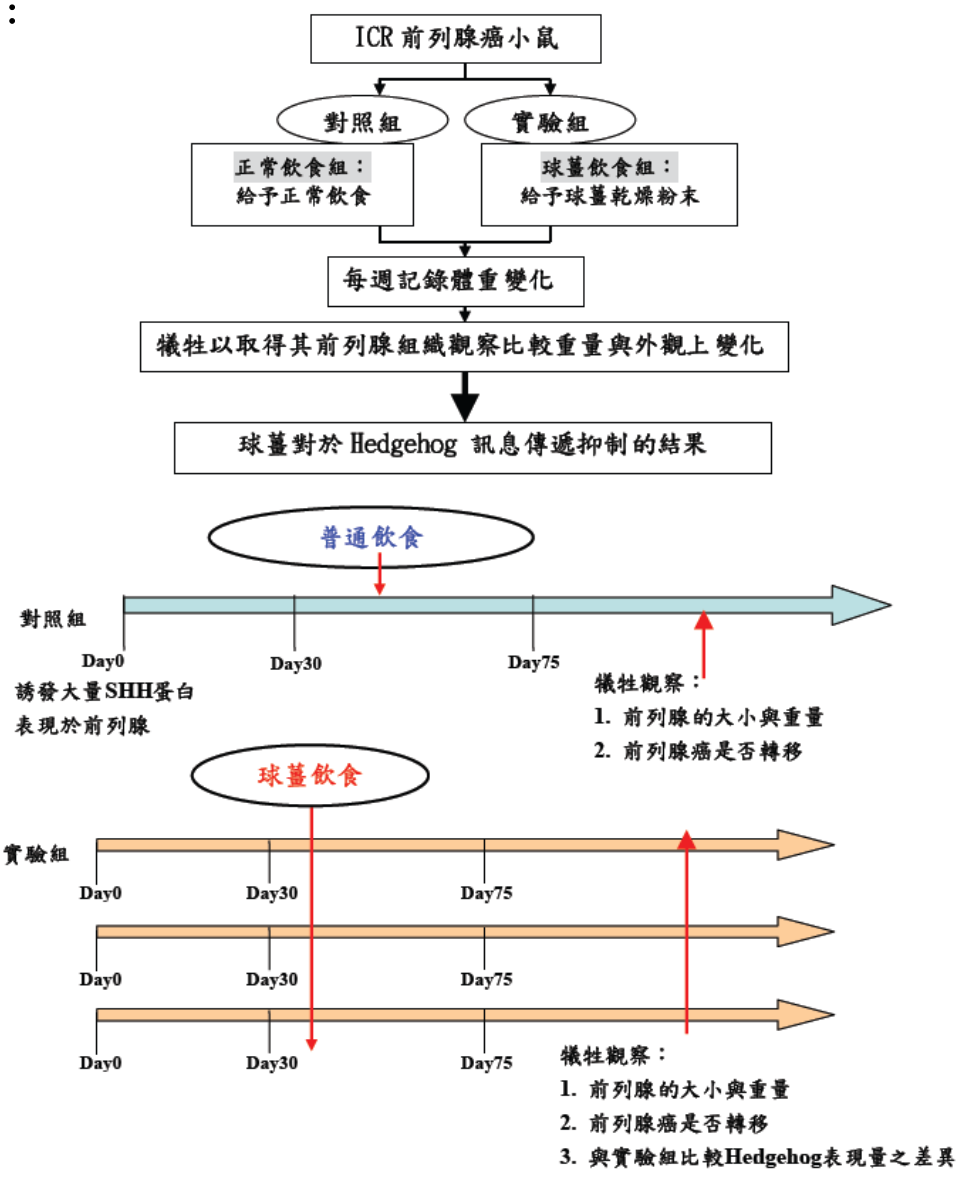
前列腺癌在西方國家一直是很棘手的疾病，十年來在美國的前列腺癌患者已經超越肺癌成為美國男性最常見的惡性腫瘤。依據衛生署統計資料顯示: 2006 年台灣前列腺癌的死亡人數佔全部癌症死亡人數 2.66%，死亡率的排行較往年增加，已達男性癌症的第 6 位。Hedgehog 的訊號傳遞已被證實與誘發前列腺癌有關。文獻指出球薑(Zingiber zerumbet)的成分對於 Hedgehog 的訊號傳遞有抑制的效果，而且球薑的成分已被運用在治療許多癌症上，然而目前還沒有文獻指出球薑的成分能夠被用來治療前列腺癌。我們實驗室之前已建立 Hedgehog 高度表現誘發小鼠前列腺癌的研究模式，因此本計畫希望能延伸先前的研究，利用小鼠模式探討誘發 Hedgehog 後，以球薑乾燥粉末餵食，觀察其組織及染色後的表現。取 50 隻的 8~10 週大的公老鼠，將老鼠先分成兩組，兩組同時打入 SHH 表現型載體；兩組在誘發後 30 天，第一組繼續採正常飲食，第二組則改為餵食球薑粉末(100ppm、250ppm、500ppm)。實驗組與對照組在 30 天及 75 天犧牲，進行取出老鼠前列腺組織，觀察並秤重後，做病理切片，以 HE 染色觀察型態上的改變，再用 IHC 染色 Gli1、Gli2、Gli3 和 p63 抗體，分析 Hedgehog 的傳遞訊號是否有減少或被抑制。如果證實在餵食球薑粉末後，可以減緩前列腺腫瘤的增生與轉移惡化，並且能夠有效地抑制 Hedgehog 訊息傳遞的路徑，對於將來臨床上前列腺癌的治療或預防，會有很大的貢獻。

關鍵詞：Hedgehog、Zerumbone、Zingiber zerumbet、Prostate cancer、GLI-mediate transcription

二、計畫緣由與目的：

實驗室一直以來持續地研究前列腺癌的發生機制與 Hedgehog 訊息蛋白所參與的角色。我們實驗室已經建立了利用 Hedgehog 誘導小鼠誘發前列腺癌的動物模式，藉以方便了解及研究人類前列腺癌的治療與預防方法。目前實驗室也發現分泌上皮細胞(luminal cells)有 CK8 的表現、基礎細胞(basal cells)有 p63 與 CK14 的表現，而且兩個細胞都有不正常增生和分化的現象。實驗室之前也萃取植物的 21 種天然多酚類化合物，並且來嘗試治療前列腺癌細胞株(PC-3)及老鼠誘發前列腺癌的試驗，由 21 種天然多酚類化合物中，尋找對 Hedgehog 訊息傳遞路徑較具專一性之萃取物。篩選後，發現 21 種天然多酚類化合物中有 2 種植物的萃取物，對於 Hedgehog 訊號抑制的能力極佳(unpublished data)。其中 Zerumbone（ZER），為可食薑 Zingiber zerumbet 的內容物，其被發現具有抗細胞不正常增殖及發炎等活性。本計畫利用球薑乾燥粉末餵食小鼠，對於 hedgehog 引發前列腺癌及前列腺癌轉移的治療是否有顯著的結果？

三、研究方法：



四、計畫成果：

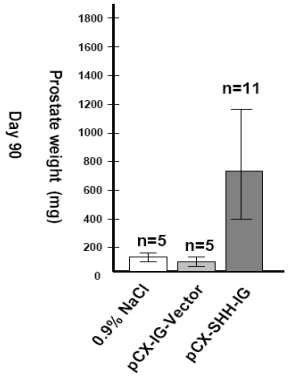


Fig 1. Prostate weigth after 90 days receiving pCX-SHH-IG

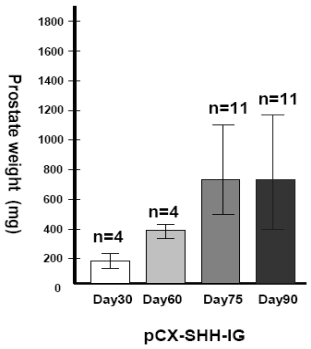


Fig 2. Prostate weight measured after 30, 60, 75 and 90 days receiving pCX-SHH-IG vectors.

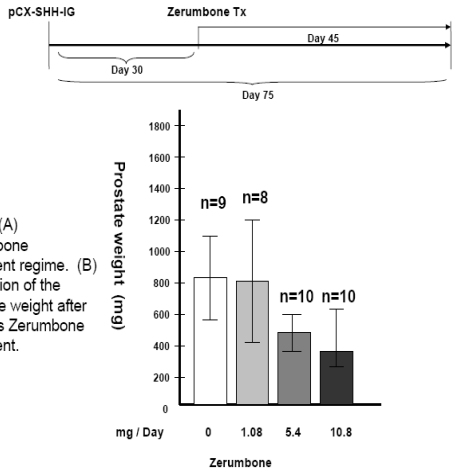


Fig 3. (A) Zerumbone treatment regime. (B) Reduction of the prostate weight after 45 days Zerumbone treatment.

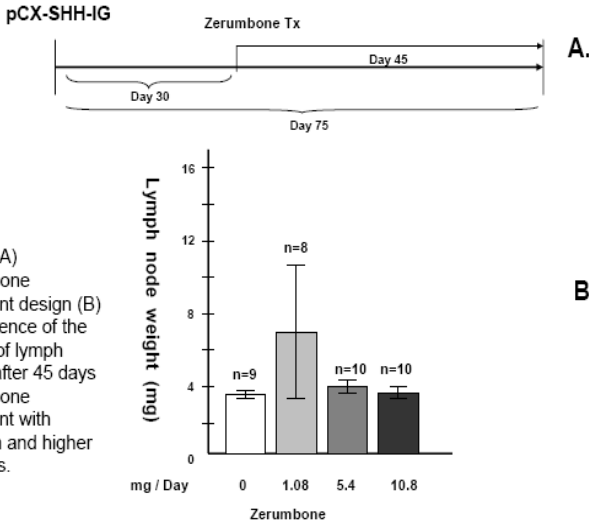


Fig 4. (A) Zerumbone treatment design (B) Consistence of the weight of lymph nodes after 45 days Zerumbone treatment with medium and higher dosages.

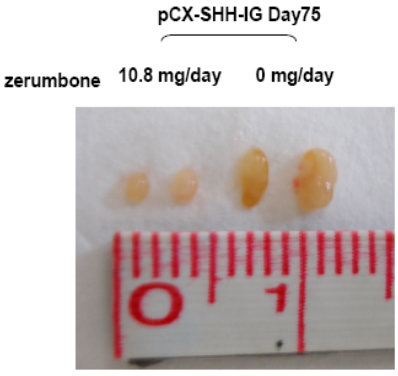


Fig 4. Inhibitory effect of zerumbone on lymph node metasis shown by the reduced size of the lymph nodes retrieved from zerumbone treated animal compared with those from none treatment group.

五、計畫成果自評：

我們製作的成果，自己相當滿意，也得到一些學界朋友的欣賞。部分作品已受邀在 2009 年展出於(Oral Presentation at International Conference and Exhibition on Nutraceuticals and Functional Foods, November 1-4, San Francisco, USA)【下圖】。但仍有可加強之處，首先是關於使用我們使用 Zerumbone 來阻斷腫瘤幹細胞(Cancer Stem cells)的效果到底如何？是否有達到預期之效果？應該要給予研究計畫的支持來進一步的分析。

以下為我參與(Oral Presentation at International Conference and Exhibition on Nutraceuticals and Functional Foods, November 1-4, San Francisco, USA)的論文

Inhibitory Effect of Zerumbone on Prostate Cancer Metastasis in a Mouse Model

Bo-Yie Chen [1], David Pei-Cheng Lin [1,2], Han-Hsin Chang [3], Yin-Ting Chen

[1] School of Optometry, Chung Shan Medical University, Taichung, Taiwan
[2] School of Medical Laboratory and Biotechnology, Chung Shan Medical University, Taichung, Taiwan
[3] School of Nutrition, Chung Shan Medical University, Taichung, Taiwan

Abstract

The purpose of this study was to examine whether the inhibitory effect of zerumbone on tumorogenesis and metasis in the prostate cancer cell line data can be applied in a mouse model. Zerumbone has been proved in cell lines to inhibit tumorogenesis through down-regulating hedgehog/Gli signaling pathway. Zerumbone was also demonstrated to inhibit the linkage between CXCR4 and CXCL12 and consequently reduced the tumor metastasis. None of these data were proved in an *in vivo* model. We have established a prostate cancer mouse model, which definitely can provide a powerful system to test these *in vitro* data. We have determined that in our animal model the prostate tumor size began to increase from day 30 after receiving hedgehog plasmid, and reached the plateau 75 days after operation. Since day 30 after SHH operation, we applied 1.08 (Low), 5.4 (Medium), and 10.8 (High) mg zerumbone respectively to animals for 45 days. Both the medium and high dosages were able to reduce the prostate tumor size. No difference of the lymph node weight was found. When metastasis sites of the other organs including the lymph node, kidney, lung, and liver were counted, we confirmed the inhibitory effect of zerumbone on metastasis. The present study was the evidence using animal model to show zerumbone effect in reducing tumorogenesis and metastasis.

Key words: Prostate Cancer, Metastasis, Zerumbone

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