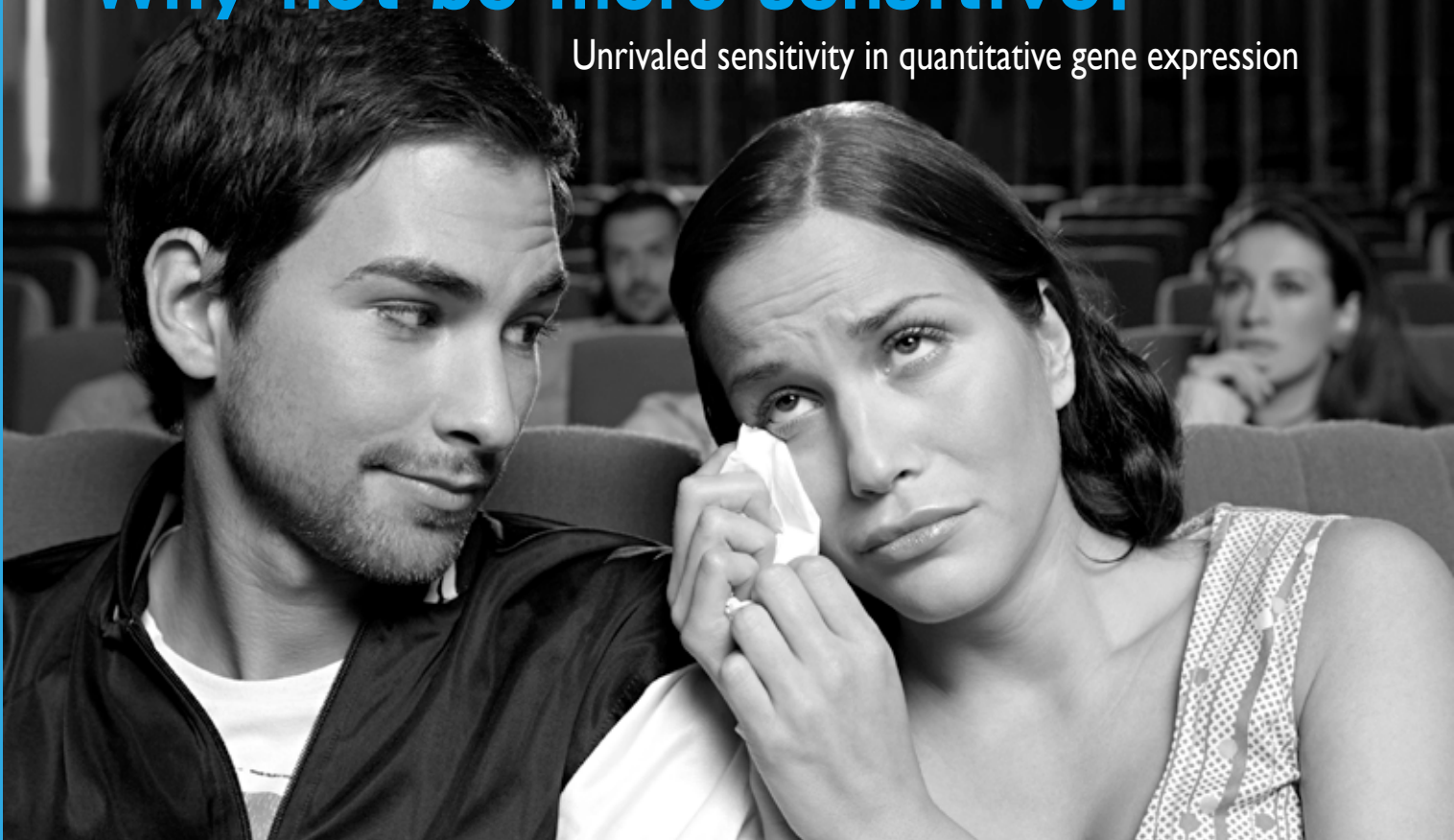
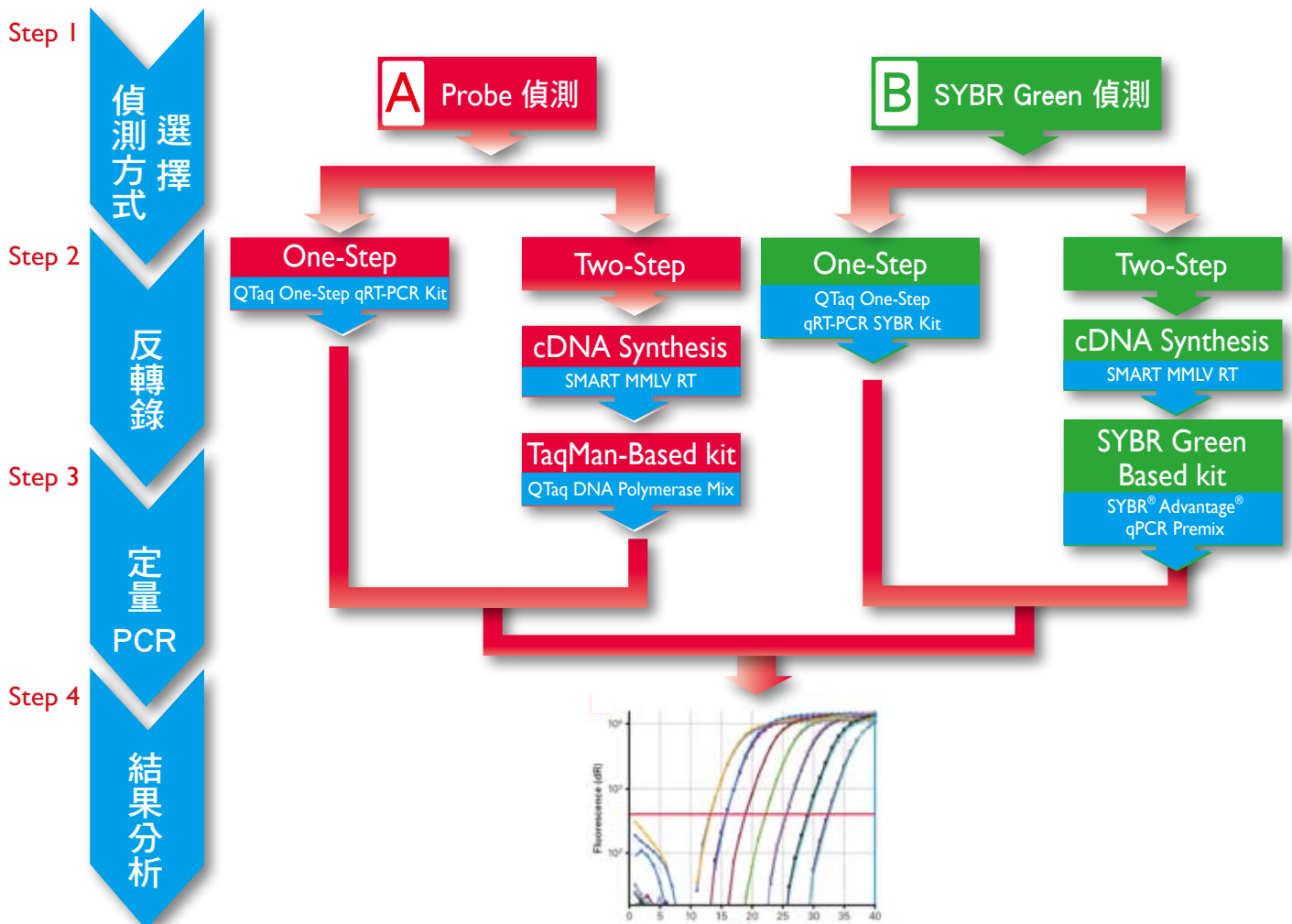


Why not be more sensitive?

Unrivalled sensitivity in quantitative gene expression



Clontech Real-Time PCR 特輯



Real-Time PCR 酵素大評比

影響即時定量 PCR 實驗的因素多，除了增強手工操作能力及儀器設備外，試劑也是一個重要的因素，但你知道如何判別試劑的品質嗎？還是就不管三七二十一的選擇儀器商所提供的試劑，卻總是苦惱著重複性不高的結果及 melting-curve 上嚴重的 multiple peaks？

如何判別試劑品質呢？

由於 Real-Time PCR 每個 cycle 都監測螢光訊息，透過擴增曲線即可直接，準確的評量試劑品質。

儀器差異-螢光的擴增曲線圖與儀器有很大關係，因此只有在同一儀器的擴增曲線圖才能反應和比較試劑的品質。

To see is to believe!

step 1.

觀察整體曲線:同一樣本，不同濃度的曲線是否平行，Negative control 的螢光基線無上揚現象。

step 2.

檢視 melting curve 可判斷專一性，同一對 primer 不同稀釋倍率的樣本應只有一個 peak。

step 3.

Ct 值與其始濃度成反比。相同的樣本，可以 Ct 值觀察出試劑的效能，Ct 值越小則效能越佳。

step 4.

絕對定量中標準曲線是建立在相同的 PCR 擴增效率，觀察曲線之間的平行性可反應出在擴增的效率，PCR 擴增的效率越相近，定量的重複性與準確性則越好。

• ROUND 1 • 專一性 (Specificity) 測試:

START!!

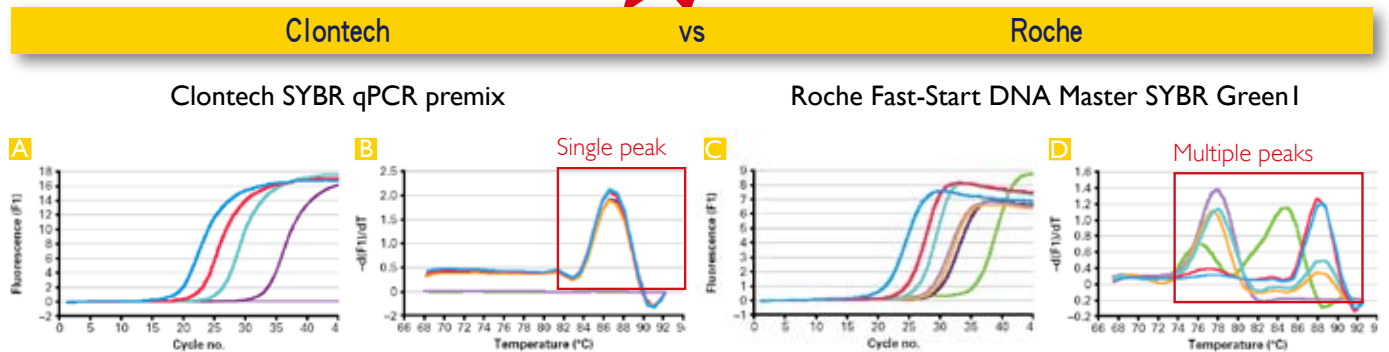


Figure 1. Reaction specificity—performance of the Clontech SYBR Advantage qPCR Premix vs. the Roche Fast Start DNA Master SYBR Green I using a Roche LightCycler. The results for the Clontech reagent are shown in Panels A and B, and those for the Roche reagent are shown in Panels C and D. The cycling conditions for the Clontech reagent consisted of 1 cycle at 95°C for 10 sec, followed by 45 cycles at 95°C for 5 sec and 60°C for 20 sec. For the Roche reagent, the cycling conditions consisted of 1 cycle at 95°C for 10 min, followed by 45 cycles at 94°C for 10 sec, 55°C for 5 sec, and 72°C for 10 sec.

• ROUND 2 • 專一性 (Specificity) 測試:

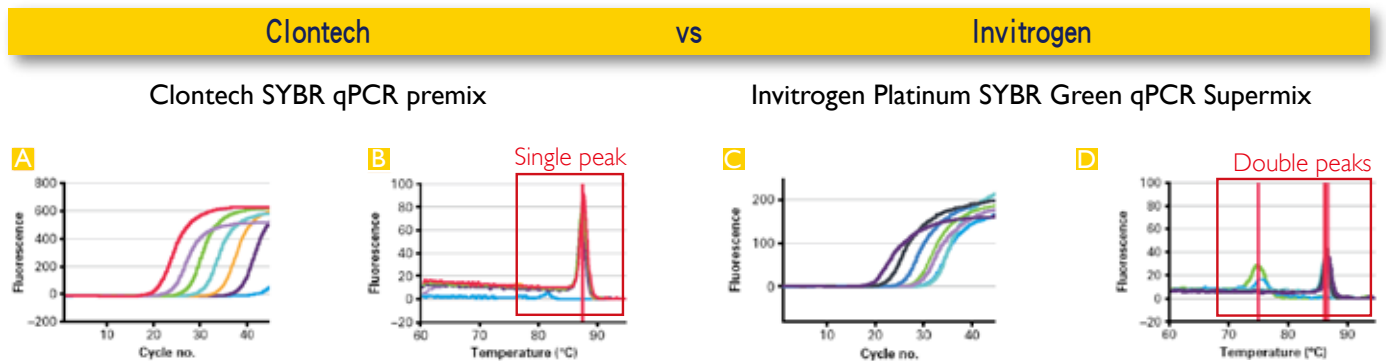


Figure 2. Reaction specificity—performance of the Clontech SYBR Advantage qPCR Premix vs. the Invitrogen Platinum SYBR Green qPCR Supermix UDG using a Cepheid Smart Cycler. The results for the Clontech reagent are shown in Panels A and B, and those for the Invitrogen reagent are shown in Panels C and D. The cycling conditions for the Clontech reagent consisted of 1 cycle at 95°C for 2 min, followed by 45 cycles at 95°C for 5 sec and 60°C for 20 sec. For the Invitrogen reagent, the cycling conditions consisted of 1 cycle at 95°C for 2 min, followed by 45 cycles at 95°C for 15 sec and 60°C for 30 sec.

• ROUND 3 • 效能(Efficiency)測試

Clontech

vs

ABI

Clontech SYBR qPCR premix

ABI SYBR Green PCR Master Mix

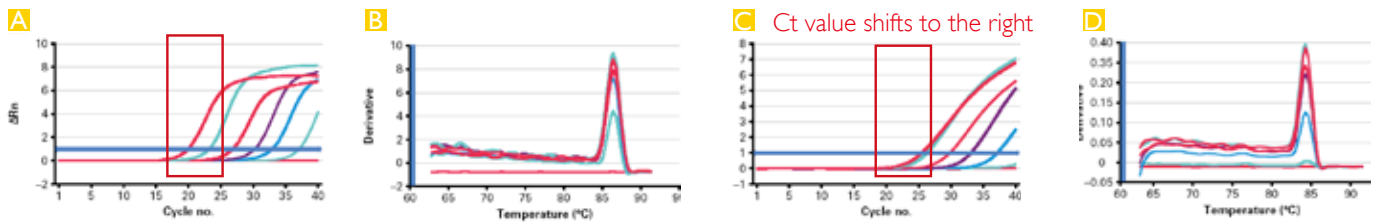


Figure 3. Amplification efficiency—performance of the Clontech SYBR Advantage qPCR Premix vs. the Applied Biosystems SYBR Green PCR Master Mix using an ABI PRISM 7000 Sequence Detection System. The results for the Clontech reagent are shown in Panels A and B, and those for the Applied Biosystems reagent are shown in Panels C and D. The cycling conditions for the Clontech reagent consisted of 1 cycle at 95°C for 10 sec, followed by 40 cycles at 95°C for 5 sec and 60°C for 31 sec. For the Applied Biosystems reagent, the cycling conditions consisted of 1 cycle at 95°C for 10 min, followed by 45 cycles at 95°C for 15 sec and 60°C for 1 min.

結果報告

1. 專一性測試結果：

在 Round 1 測試中，觀察整體擴增曲線，Clontech 螢光曲線整體平行一致，Roche 的低濃度 template 螢光基線則有微微上揚。比較 melting curve，Roche 出現 multiple peaks，在低濃度的 template 此狀況更為明顯，Round 2 測試中 Invitrogen 的 melting curve 則出現了 double peaks，結果顯示 Clontech 酵素的專一性較佳，輕輕鬆鬆打敗 Invitrogen 跟 Roche。

2. 效能測試結果：

在 Round 3 測試中，觀察整體擴增曲線，ABI 酵素的 Ct 值明顯比 Clontech 要高，而使用 Clontech 的試劑則整整比 ABI 減少將近一半的時間（50 mins vs 90 mins），結果顯示，Clontech 擁有較佳的酵素效能。

輕輕鬆鬆贏得
Real-Time PCR
試劑大賞!!

答案是

SYBR Advantage qPCR Premix

產品特色

高靈感性-

只要 5 copies 的 template 即可測得，勝於其它廠牌

高專一性-

含 Hot start antibody 的 Full-length Taq polymerase，減少非 PCR 產物

方便使用-

2X SYBR Green I master mix

適用於多種 Real-Time PCR 機器-另外附 Reference ROX dye (LCM/LED)

Clontech April 2006 14-15

www.clontech.com

SYBR Advantage qPCR Premix Outperforms Competitors

Clearly, the Clontech SYBR Advantage qPCR Premix outperforms much of the competition in terms of specificity (single product as judged by melting curves), efficiency (amplification plots [shown] and standard curves [not shown]) and sensitivity (lower Ct values). Moreover, the overall

time for the qPCR analysis can be shortened dramatically—nearly in half compared to the times for other qPCR chemistries—when using the Clontech SYBR Advantage qPCR Premix.

— Clontech Laboratories, Inc

誰能跟我比



備註: 25 uL / reaction

Cat.#	Product	Package	促銷價
639676	SYBR ADVANTAGE qPCR PREMIX	200 rxns	\$4,360.

QTag™ One-Step qRT-PCR Kit

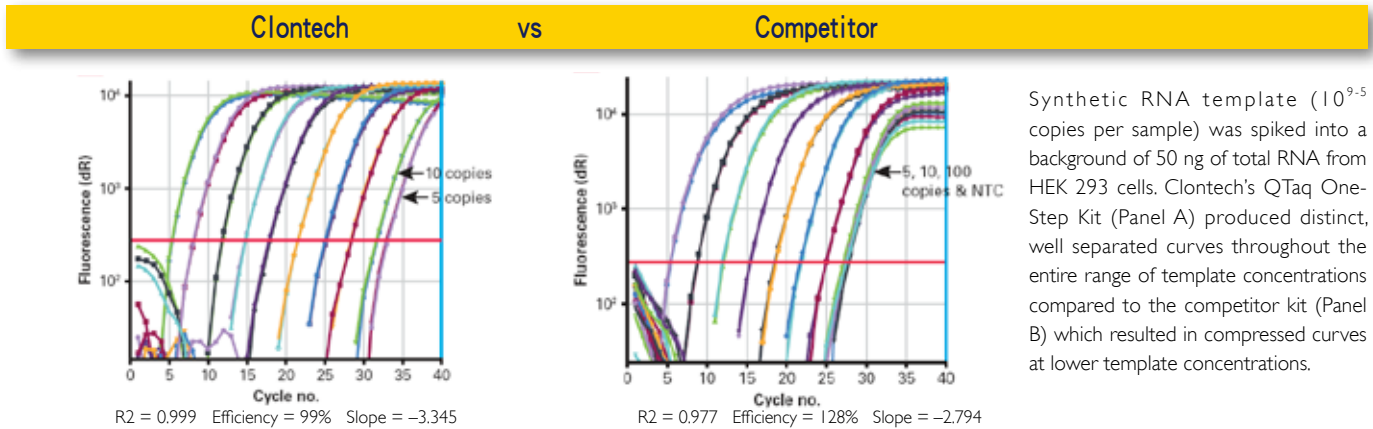
您了解您使用的酵素偵測範圍及靈敏度嗎?

產品特色

最方便：只要一個反應管，一個步驟，減少 cross-contamination 的風險

最靈敏：只要5個 copies 的 template 就可偵測

最實用：可應用於 One-step 或 Two-step



Synthetic RNA template (10^{9-5} copies per sample) was spiked into a background of 50 ng of total RNA from HEK 293 cells. Clontech's QTag One-Step Kit (Panel A) produced distinct, well separated curves throughout the entire range of template concentrations compared to the competitor kit (Panel B) which resulted in compressed curves at lower template concentrations.

限時特價

備註: 25 uL / reaction

Cat.#	Product	Package	促銷價
639518	QTag One-Step qRT-PCR SYBR Kit	200 rxn	\$10,800.
639519	QTag One-Step qRT-PCR SYBR Kit	1000 rxn	\$51,840.
639520	QTag One-Step qRT-PCR Kit	200 rxn	\$10,800.
639521	QTag One-Step qRT-PCR Kit	1000 rxn	\$51,840.

每一包裝含: 1. 60X qRT Mix 2. 50X QTag™ DNA Polymerase Mix 3. 2X One-Step qRT-PCR Buffer plus / without SYBR

SMART™ MMLV RT

產品特色

高靈敏度- 5個 copies 的 RNA 即可

合成長度達 10 kb

保留 RNase H 活性，提高 cDNA 產量

嚴謹的 QC -給你真正 RNase-free 及 DNase-free 的 RT

RT 效果不好?
是否使用的酵素問題?

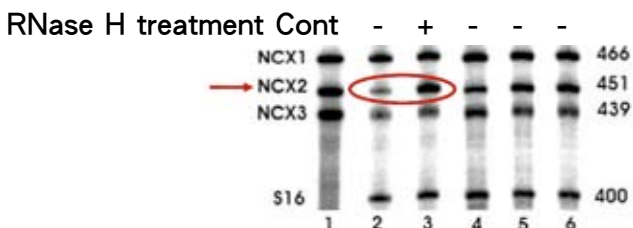
Clontech 最嚴謹的 QC 給你不高鬼的 RT!

Vendor	1st Strand	Ribonuclease test	Endonuclease test	Exonuclease test	DNA Contam. (Bacterial)	In process QC
Clontech	Y	Y	Y	Y	Y	Y
Promega	Y	Y	Y			Y
Ambion	Y		Y	Y		Y
NEB	Y	Y		Y		Y
Stratagene		Y	Y	Y		

反轉錄的 cDNA 經 RNase H 處理後，增加了 PCR 反應的靈敏度。

Denatured time (min) 1 5

Denatured temperature 94 97 100



"We show that the template of one gene of a multigene family becomes more readily available for PCR amplification only after RNase H treatment, while two other genes are not affected by RNase H treatment."

BioTechniques 32: 1224-1225 (June 2002)

你的 RT 有RNase 嗎?

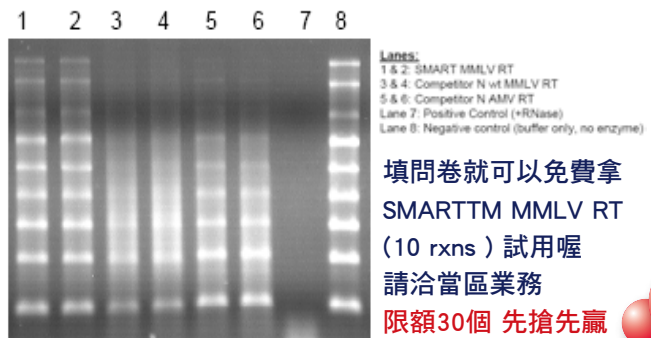


Figure 1. SMART MMLV RT has less RNase activity than the competition. An RNA Ladder was incubated with 200 units of either SMART MMLV RT (Lanes 1 & 2), or another supplier's MMLV RT (Lanes 3 & 4) and AMV RT (Lanes 5 & 6) at 37°C for 1 hr; and then analyzed by formaldehyde/agarose gel electrophoresis. Lane 7: positive control; Lane 8: negative control (buffer only, no enzyme).

填問卷就可以免費拿
SMART™ MMLV RT
(10 rxns) 試用喔
請洽當區業務

限額30個 先搶先贏

