

食物致癌物和其代謝物之遺傳毒性

李　輝

中山醫學院生化科主任

簡介

最近發現有許多異環狀胺類由烹調食品和加熱過程中形成，這些化合物在Ames test上都被證明是致突變物。kasai（1980）[1]首先由烤沙丁魚的烤焦表面抽取到IQ化合物，它在PCB誘發的老鼠肝臟微粒體酵素活化作用下成為一對TA98具有強烈致突變性的化合物。IQ也能引起老鼠肝臟培養細胞的不按計劃的DNA合成（unscheduled DNA synthesis）。它的代謝產物能和DNA共價結合。在體內的實驗方面，IQ被證明能引起老鼠肝癌以及其他癌症。這類食物中的致突變物是否亦能引起人類的癌症，到現今我們還不明瞭，可是由流行病學的數據顯示食物是引起人類癌症的主要原因（Hatch et al., 1983）[2]，因此了解這類食物致突變物在體內代謝的途徑以及在不同種類動物和器官是否有差異性，是明瞭這類食物致突變物對人體影響的一種可行方法。

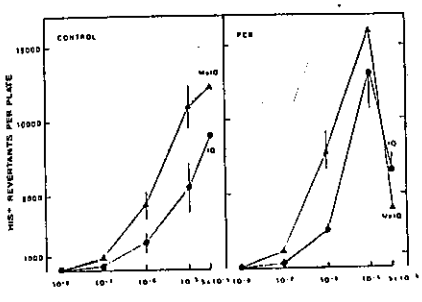
雖然IQ類化合物在Ames test上有很強的致突變性，但是在許多其他短期遺傳毒性測定方法上卻是很弱的遺傳毒物。因此須要由人類細胞或其他哺乳類細胞來測定它們的遺傳毒性。

為了了解IQ化合物對體內的影響，許多學者以老鼠肝臟微粒體酵素、肝臟細胞以及人類肝臟微粒體酵素來代謝活化IQ化合物，並以放射性追踪法探討它的代謝途徑和代謝物的化學構造。更進而到代謝物和DNA互相作用的分子層次，也分離純化出它們的DNA adduct在IQ化合物的致癌機制上已有相當深入的探討，茲將最近幾年來有關這方面的研究加以整理，敘述如後。

IQ類化合物在哺乳細胞的遺傳毒性

IQ類化合物在Ames test上對細菌是超級強烈的致突變物早在1980年以後即被知道，至於對真核細胞的哺乳類細胞是否亦能造成遺傳毒性則在最近數年才開始有學者研究。Holme（1988）[3]曾比較IQ和MeIQ在沙門氏桿菌和哺乳細胞上的遺傳毒性，因為IQ是間接致突變物，須要肝臟酵素代謝活化始為致突變物，因此作者則用分離培養的老鼠肝臟細胞在代謝活化IQ和MeIQ來測定它的代謝物對TA 98的致突變性，結果如Fig. 1，亦能形成線性濃度關係。在不按計畫的DNA合成的實驗上，MeIQ能引起最強的UDS如Fig. 2所示。若以Alkaline elution的方法來測定IQ類化合物對V_{79}細胞DNA的損傷性，由Fig. 3的數據顯示MeIQ較IQ對DNA的損傷為大，但都須要PCB誘發的老鼠肝細胞活化才有作用。可是對照組的MNNG則無須肝細胞活化就能引起DNA損傷。

Fig. 1

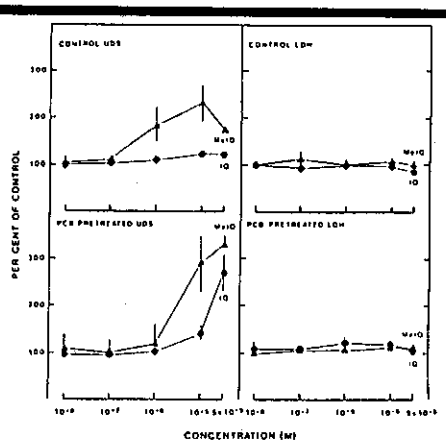


Concentration dependence of IQ and MeIQ induced mutations in *Salmonella typhimurium* with hepatocyte activation. 2 h after plating 1.5×10^6 hepatocytes isolated from untreated or PCB-pretreated rats were co-cultured with *Salmonella typhimurium* TA98 for 1 h as described in Materials and Methods. Values are means ± SD (bars) of 3 separate experiments.

在誘發SCE和6-TG′突變種（V_{79}）的遺傳毒性測定上，若沒有肝細胞的活化，IQ和MeIQ都不能引起V_{79}細胞產生SCE頻率較控制組為高，但是若經過PCB處理的肝細胞活化則IQ和MeIQ能誘發2倍以上的SEC頻率，且和處理的

濃度呈一遞增關係，如Table 1和2所示。但是IQ和MeIQ在6-TG' 突變種的誘發上，即使經過肝細胞的活化仍然僅有少數的突變種形成，數據如Table 3所列。

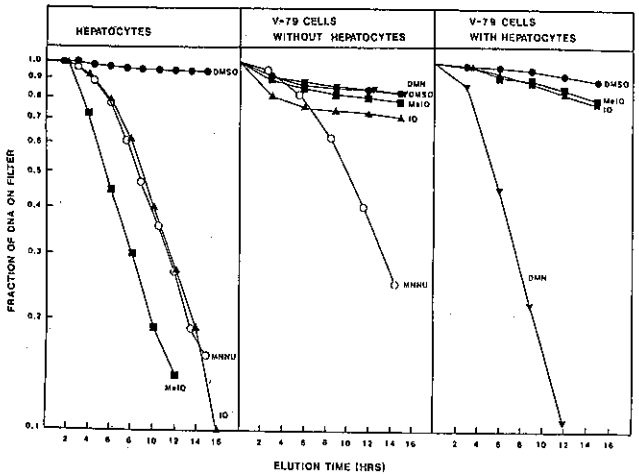
Fig. 2



Concentration dependence of IQ and MeIQ induced unscheduled DNA synthesis in monolayer cultures of hepatocytes. Monolayers of hepatocytes isolated from untreated or PCB-pretreated rats were exposed to various concentrations of IQ and MeIQ in Hank's-Hepes buffer. After a 3-h exposure period, the hepatocytes were changed back to ordinary cell culture medium containing hydroxyurea and ^{3}H-TdR. Incorporation of ^{3}H-TdR into nuclear DNA was measured after a total incubation time of 20 h. Cytotoxicity was determined by LDH leakage 20 h after plating. Values are means ± SD (bars) of 3 separate experiments.

Minkle和Carrano（1984）[4]亦曾報告IQ在老鼠骨髓細胞實驗上不會引起染色體變異，但是會引起SCE頻率增高，所以IQ雖然在細菌的實驗上是很強的致突變物，但是在大多數哺乳類細胞的生物檢定都證明是很弱的遺傳毒物。這兩者間差異造成的原因有許多解釋，有人認為IQ的代謝物無法和哺乳細胞的DNA發生作用，或許是因為它的滲透性太低，穩定性不夠，或是其代謝物很快的被解毒，在Holme的實驗裏IQ能被老鼠肝細胞活化而非解毒反應，而且能通過哺乳類細胞的細胞膜而不會造成傷害，進而發現IQ和MeIQ在肝細胞代謝活化的代謝物半生期有45分鐘之

Fig. 3



Studies of IQ and MeIQ induced DNA damage in isolated hepatocytes and V79 cells measured by alkaline elution. DNA damage of hepatocytes from PCB-pretreated rats incubated as cell suspensions, was measured after exposure to the test substance (10^{-4} M IQ/MeIQ; 0.3 mM MNNU) for 3 h. Monolayers of V79 cells co-cultured with or without PCB-pretreated rat hepatocytes were exposed to the test substance (0.5 mM IQ/MeIQ; 0.3 mM MNNU; 3 mM DMN) for 2 h. The data shown are from representative experiments.

TABLE 1

EFFECT OF IQ, MeIQ AND CYCLOPHOSPHAMIDE (CPA) ON INDUCTION OF SCEs IN V79 CELLS INCUBATED WITH OR WITHOUT RAT HEPATOCYTES

Test substance (μM)		Number of SCEs per chromosome	
		Without hepatocytes	With hepatocytes
DMSO		0.30	0.28
IQ	100	0.30	0.37
MeIQ	100	0.30	0.47
CPA	200	0.26	5.80

Chinese hamster V79 cells, incubated with or without PCB-pretreated rat hepatocytes, were exposed to test substance for 2 h and then further incubated for 30 h. Values are means of at least 300 scored chromosomes from 1 Expt. (for mean ± S.D. see Table 2).

TABLE 2

CONCENTRATION-DEPENDENT INDUCTION OF SCE IN V79 CELLS COCULTURED WITH RAT HEPATOCYTES

Test substance (μM)		Number of SCEs per chromosome
DMSO		0.29 ± 0.03
IQ	50	0.45 ± 0.12
	100	0.53 ± 0.14
	500	0.70 ± 0.17
MeIQ	50	0.39 ± 0.02
	100	0.41 ± 0.06
	500	0.68 ± 0.10

Chinese hamster V79 cells incubated with PCB-pretreated rat hepatocytes were exposed to test substance for 2 h and then further incubated for 30 h. Values are means ± SD of 3 separate Expts.

長。因此有IQ化合物在哺乳類細胞遺傳毒性較細菌為低仍須由IQ和MeIQ的代謝途徑進一步明瞭才可以圓滿解釋。

Aeschbacher和Ruch（1989）[5]用人類淋巴細胞來測定IQ和MeIQx的遺傳毒性。由Table 4所知IQ和MeIQx在很高的濃度下，仍然無法造成人類淋巴細胞的染色體變異。在SCE測定上，濃度達到800ng／ml的IQ和MeIQx，在沒有S_9狀態下，也無法引起SCE頻率增高，若在S_9活化下，則僅有統計上（$P<0.05$）的SCE增加的意義，結果如Table 5，6所示。由這個實驗可知IQ和MeIQx對人類淋巴細胞僅能在有S_9活化下，能引起SCE的少量增加，這表示

TABLE 3

EFFECTS OF IQ AND MeIQ ON THE FREQUENCY OF 6-TG^r MUTANTS IN CHINESE HAMSTER V79 CELLS CO-CULTURED WITH PCB-PRETREATED RAT HEPATOCYTES

Test substance (μM)		6-TG^r mutants per 10^6 cells			
		Expt. 1	Expt. 2	Expt. 3	Expt. 4
DMSO		< 0.7	2.2	4.3	7.5
IQ	100	5.1	–	10.8	7.4
	250	–	–	4.9	7.6
	500	–	4.7	–	–
MeIQ	100	1.4	–	9.6	5.1
	250	–	–	5.0	10.7
	500	4.6	3.4	–	6.9
DMBA	7	–	–	–	24.0
MNNU	100	27.6	–	10.2	–

V79 cells co-incubated with PCB-pretreated rat hepatocytes for 24 h and processed according to Jenssen et al. (1984). 500 μM IQ/MeIQ gave a 50–80% reduction of plating efficiency.

TABLE 4

Evaluation of IQ and MeIQx for chromosome aberrations in human lymphocytes

μg/ml culture	IQ +S9			MeIQx +S9		
	Gaps	Breaks	Other	Gaps	Breaks	Other
0	1.5	1.5	0	2.5	2.0	0
0.4	2.0	2.0	0	2.0	1.0	0
0.8	1.0	3.5	0.5	1.5	1.0	0
5.0	1.5	1.5	1.0	1.5	2.5	0
25.0	1.5	2.0	0.5	1.0	1.5	0.5
50.0	1.0	3.5	0	2.0	0.5	0
100.0	1.5	1.0	1.5	1.5	1.0	0
250.0	3.5	3.0	0	1.5	1.5	0
500.0	0.5	4.0	0.5	2.0	0.5	0.5
1000.0	3.0	2.0	0	1.5	2.0	0.5
Endoxan 30 μg	2.0	18.0	12.0	2.5	23.0	13.5

The values represent the number of metaphases with at least one gap, or one break or one other aberration (e.g. translocation) per 100 metaphases. The mean of two times 100 metaphases is given. Statistical evaluations were carried out and overall standard errors of the mean were 0.30 for IQ, 0.23 for MeIQx and 0.34 for DIFCO when the sum including all types of aberrations were considered. Results given in the tables with the presence of S9 only. A few selected high doses for IQ and MeIQx were also tested without S9. The total number of gaps, breaks and other aberrations were 4.0 or 4.5 for 1000 μg/ml, 5.0 or 3.5 for 500 μg/ml and 4.5 or 6.5 for 250 μg/ml culture for IQ and MeIQx respectively (IQ = 4.0 and MeIQx = 4.5 for negative controls).
For IQ and MeIQx the solvent control (0 μg/ml culture) consisted of 0.3 ml H_2O/5 ml culture.
For DIFCO beef extract (basic fraction) mg equivalents (mgE) of the 'fresh' product are given. 0 mgE/ml culture corresponds to 0.3 ml DMSO 25% (solvent control).

IQ化合物對DNA的損傷力主要是點突變而非構造破壞。造成DNA之SCE頻率升高可能是修復機制改變和基因突變同時發生，使得DNA無法迅速修復，這也可以來解釋為IQ化合物是低強度的致癌物。

另外人類細胞具有很強的修復DNA系統和高活性的解毒酵素，因此可以減低對人類細胞的遺傳毒性，況且在細菌體中有很高的O-acyltransferase活性，能很快的將IQ化合物轉變為致突變物，所以在細菌的生物檢定上是很強的致突變物，而在哺乳類細胞則為弱的遺傳毒物。

IQ類化合物的代謝途徑

Mc Manus（1988）[6]以人類肝臟微粒體酵素代謝活化IQ化合物，這些人類肝臟是由六個死亡的病人，在30分鐘—60分鐘之間取出肝臟，再以刀切取4立方公分的肝組織放在液態氮中冷凍，然後移到－80℃的低溫冷藏箱貯存。人類肝臟細胞的微粒體和S_9fraction則以McManus（1984）[7]的方法抽取之。IQ化合物經過這六個人的肝臟酵素活化後，以Ames test TA98測定其致突變性，由11830±320到42830±290，相差四倍左右。由Fig. 4、5、6顯示人類肝臟酵素活化IQ化合物的最適宜反應狀態。同時他以2-AF做為標準樣品作一比較。IQ的致突變性以pre incubation方法測定。人類肝臟酵素能增強IQ致突變性幾近200倍。例如10μg IQ在標準測定法可產生4000個反突變菌落數，這個數字在preincubation方法僅要0.05μg IQ即可達到。所以在做promutagen致突變性測定時大多採用preincubation方法。

Fig. 5的結果顯示微粒體蛋白質和S_9蛋白質濃度對IQ致突變性的影響，在0.1mg的微粒體蛋白質和1mg的S_9蛋白質濃度範圍內，其引起的反突變菌落數呈線性關係。當比較六個不同來源的微粒體酵素，對IQ和AF的代謝活化的影響如Fig. 7所示。大概有4～2倍的差異。肝臟中的Glutathione是最主要的nucleophil，能去除electrophils而抑制IQ和AF的致突變性30和57%，如Fig. 8所示。作者另取α—naphthoflavone和metyrapone兩種抑制P-450酵素活性的抑制劑，來研究是否能抑制IQ的致突變性，由Fig. 6可知α—naphthoflavone分別在0.1μm和50μm能抑制IQ和AF的致突變性93%和80%，而Metyrapone對AF無作用，但是對IQ則有72%的抑制性。由以上的實驗我們可以知道人

TABLE 5

Evaluation of IQ and MeIQx for SCE in human lymphocytes in culture

μg/ml culture	IQ SCE/cell ± SEM		MeIQx SCE/cell ± SEM	
	−S9	+S9	−S9	+S9
First experiment				
0	5.4 ± 0.3	4.5 ± 0.2	5.2 ± 0.2	5.4 ± 0.2
0.1	5.0 ± 0.3	4.6 ± 0.3	6.2 ± 0.4	7.2 ± 0.5
0.2	5.2 ± 0.3	5.5 ± 0.3	6.4 ± 0.3	6.6 ± 0.4
0.4	5.0 ± 0.3	5.2 ± 0.2	6.8 ± 0.4	7.3 ± 0.5
0.6	5.8 ± 0.3	6.0 ± 0.3	6.2 ± 0.3	8.2 ± 0.4
0.8	5.2 ± 0.3	5.1 ± 0.3	6.0 ± 0.4	7.7 ± 0.4
Thiotepa 0.3 μg	13.6 ± 0.5		17.6 ± 1.2	
Endoxan 2.5 μg		12.7 ± 0.6		15.5 ± 0.7
Second experiment				
0	4.6 ± 0.2	4.7 ± 0.2	4.2 ± 0.2	4.6 ± 0.2
25	5.4 ± 0.2	10.6 ± 0.5	5.0 ± 0.2	6.6 ± 0.6
50	4.7 ± 0.2	10.0 ± 0.4	5.4 ± 0.3	6.5 ± 0.4
75	5.6 ± 0.3	11.7 ± 0.5	5.5 ± 0.3	7.1 ± 0.3
100	5.1 ± 0.3	11.4 ± 0.6	5.8 ± 0.3	6.6 ± 0.3
250	5.7 ± 0.3	−	6.0 ± 0.3	−
Thiotepa 0.3 μg	12.3 ± 0.4		15.5 ± 0.5	
Endoxan 2.5 μg		10.5 ± 0.4		17.4 ± 1.0

The values of the first experiment represent the mean number of SCE per 30 cells ± SEM. Each compound was tested in double culture hence 2 × 30 metaphases were evaluated both in the presence (+S9) and the absence (−S9) of liver enzyme metabolizing system. For the second experiment (IQ and MeIQx) 2 × 40 metaphases were evaluated in double cultures and results were expressed as for experiment 1.
For the negative controls the respective solvents were added to the cultures: 0.1 ml H_2O for IQ, 0.3 ml H_2O for MeIQx and 0.2 ml 25% DMSO for DIFCO. For 250 μg of MeIQx and IQ (+S9) evaluation was not possible due to a cytotoxic effect.

TABLE 6

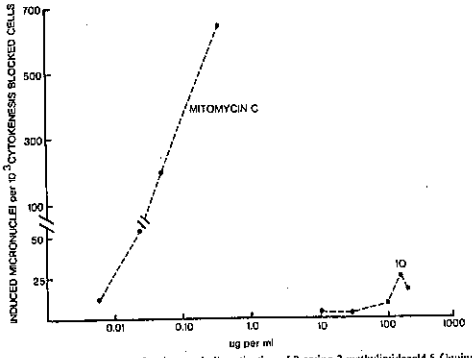
Statistical significance values

			P values for SCE	
			Concentration versus control	Linear trend
IQ	0.1–0.8 μg/ml	−S9	0.919	0.439
		+S9	0.001	0.006*
	25–250 μg/ml	−S9	0.105	0.061
		+S9	0.000	0.000*
MeIQx	0.1–0.8 μg/ml	−S9	0.005	0.307
		+S9	0.000	0.000*
	25–250 μg/ml	−S9	0.000	0.000*
		+S9	0.000	0.000*
DIFCO	5–200 gE/ml	−S9	0.048	0.000(*)
		+S9	0.000	0.000*

*Results were significantly different to the solvent control and also gave a significant linear trend.
(*)Borderline case.
Values $P < 0.05$ were considered as significant, 0.000 indicates high statistical significance. The statistical evaluations were carried out on transformed data (Poisson distribution). Two tests were carried out using a one-way analysis of variance. One to compare the treatment group to the solvent control and the other to calculate the linear trend. Since the three test substances did not significantly induce CA, no statistical values were given for CA.

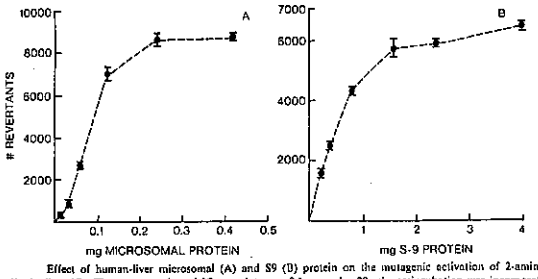
類肝臟微粒體酵素活性較PCB誘發的老鼠肝臟酵素差，但較猴子的高20倍。同樣的結果也由Dolora（1980）[8]以四個人的肝臟酵素測定細菌用牛肉汁抽取物致突變性的實驗得到。

Fig. 4



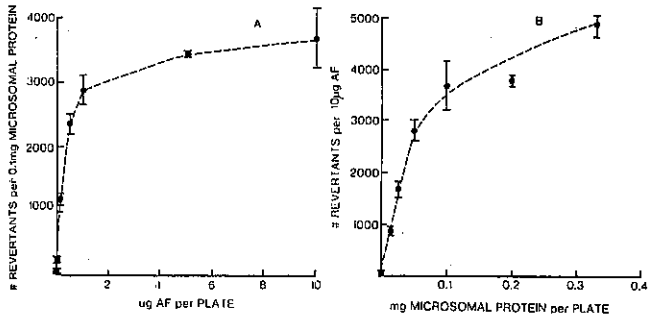
Dose-response curve for the metabolic activation of 2-amino-3-methylimidazo[4,5-*f*]quinoline by human-liver microsomes to a mutagen in the absence (A) and presence (B) of a preincubation step in the Ames test. The human-liver protein concentration was 0.1 mg per plate. Results represent $x \pm SD$, $n = 3$.

Fig. 5



Effect of human-liver microsomal (A) and S9 (B) protein on the mutagenic activation of 2-amino-3-methylimidazo[4,5-*f*]quinoline (IQ). The concentration of IQ per plate was 0.1 μg and a 20 min preincubation was incorporated into the Ames test. Results represent $x \pm SD$, $n = 3$.

Fig. 6

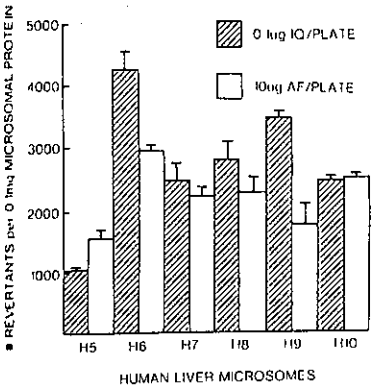


2-Aminofluorene (AF) mutagenicity in vitro as a function of substrate concentration (A) and human-liver microsomal protein concentration. The concentration of microsomal protein in (A) was 0.1 mg and the concentration of AF in (B) was 10 μg per plate. Results represent $x \pm SD$, $n = 3$.

代謝活化異環狀胺類化合物的第一個反應是將胺基N-hydroxylation，Mulder（1982）[9]即確定兩種Glutathione結合的N-hydroxy-α-aminoflurorene，Mc Manu（1988）[6]的實驗證明Glutathione能抑制IQ和AF的致突變性，但是在preincubation混合液中過多的Glutathione仍然無法做一個有效的異環狀胺類代謝物的清除者，因此作者認為人類肝臟微粒體酵素系統中可能含有Glutathione和Glutathione transferase，所以其代謝活化性較老鼠的低。

α—naphthoflavone能抑制IQ的致突變性，即表示cytochrome P-450以多環芳香羥形式存在於肝臟微粒體酵素系統中，但是α—naphthoflavone對不同來源的酵素系統的作用程度並不相同，同時對不同的致突變物的代謝物的作用亦不同，這種現象反應了cytochrome 450可能含有不同異構酶。

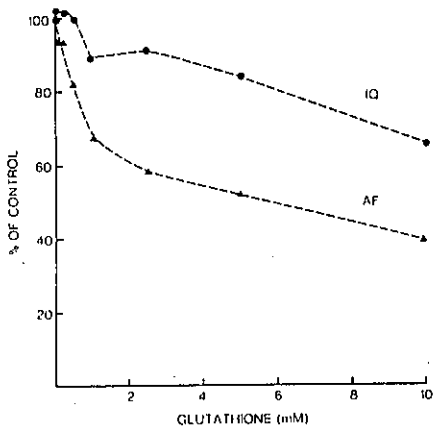
Fig. 7



Mutagenic activation of 2-amino-3-methylimidazo[4,5-*f*]quinoline (IQ) and 2-aminofluorene (AF) by human-liver microsomes from 6 subjects. The microsomal protein concentration was 0.1 mg per plate and IQ and AF concentrations were 0.1 μg and 10 μg per plate, respectively. Results represent $x \pm SD$, $n = 3$.

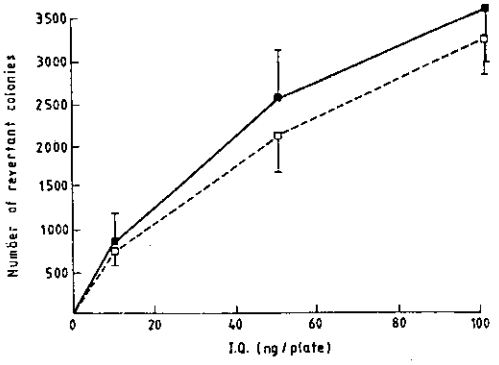
IQ的代謝活化過程中Sulphation沒有參與，這個結果是由Paterson和Chipman（1987）[10]將培養細胞的培養液中的Sulfate除去後和控制組比較，對TA98的反突變菌落數並沒有顯著改變，如Fig. 9所示。後來他們又用兩種Sulphotransferase抑制劑：PCP和2.6-DCNP進一步實驗證明IQ在代謝活化過程中Sulphation並沒有參與。結果PCP能有效抑制IQ的致突變性，而2.6-DCNP則無此作用（Table 7）。在1983年Nagao[11]等也認為PCP能抑制IQ的致突變性。可是在Paterson和Chipman的實驗裏又發現沒有Sulphate和加入過量Sulphate都對IQ的致突變性沒有作用，而且加2.6-DCNP也沒有影響。IQ的代謝物在TA98／1,6 DNP$_6$菌株沒有致突變性（Fig. 10）。雖然許多實驗都證

Fig. 8



Effect of glutathione on the mutagenic activation of 2-amino-3-methylimidazo[4,5-*f*]quinoline (IQ) and 2-aminofluorene (AF) by human liver microsomes. The incubation mixture and conditions of the assay are described under Methods.

Fig. 9



Mutagenicity of IQ towards *S. typhimurium* TA98 following a 2-h pre-incubation assay with isolated rat hepatocytes (1×10^6/plate). Incubations were performed in the presence (▽) or absence (■) of inorganic sulphate. Results are shown as mean number of revertants ± SEM for four hepatocyte preparations. The frequency of mutation with or without sulphate is not significantly different at any dose level of IQ examined.

明芳香胺類在活化過程中都有Sulphation參與在N-hydroxy-lation的過程中，但是由以上的結果可推論IQ的代謝活化過程裏Acetylation較Sulphation為重要。

TABLE 7

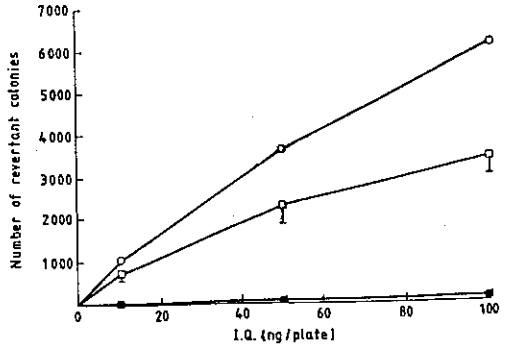
Effect of PCP and 2,6-DCNP on IQ mutagenicity towards *S. typhimurium* TA98 in the presence of isolated rat hepatocytes

Addition	Number of revertants per plate (minus spontaneous revertants)
DMSO	876 ± 82
PCP + DMSO	413 ± 61*
2,6-DCNP + DMSO	648 ± 117

Salmonella/hepatocyte assays were performed in the absence or presence of PCP or 2,6-DCNP (10 μM) added in DMSO (20 μl). Pre-incubation with hepatocytes (1.0×10^6) was for 2 h at 37°C with 10 ng IQ. Results are the mean ± SEM for five hepatocyte preparations each analysed in duplicate. Where indicated (*) the value is significantly different ($P < 0.01$; unpaired *t*-test) from control incubations (addition of DMSO alone). At these concentrations of inhibitors, no toxicity to Salmonella or hepatocytes was observed. The spontaneous reversion rate was <30 colonies/plate throughout.

至於MeIQx的活化酵素仍然還是存在於微粒體中，若以3-methylcholanthrene（3-MC）和PCB誘發的肝臟微粒體酵素系統活化MeIQx的能力較PCB誘發的為高。Yamazoe

Fig. 10



Mutagenicity of IQ towards *S. typhimurium* TA98 (□, ▽) and TA98/1,8DNP$_6$ (■) following a 2-h pre-incubation assay using hepatocytes from control rats (□, ■) and Aroclor 1254-induced rats (○). Results are shown as the mean number of revertants ± SEM for six hepatocyte preparations except in the case of Aroclor-induced preparations (mean of two). The number of hepatocytes used per incubation was 1.0×10^6.

（1988）[12]為了要了解其原因就將Cytochrome-P-450純化分出五種形式的酶系統，研究它們活化MeIQx的能力，如Table 8可知，3-MC誘發的酵素較沒有誘發的控制組之酵素活化MeIQx的能力高9倍，PCB誘發的則高達20倍，當加入P-448形式的Cytochrome P-450的抑制劑flavone和benzo-flavone則反突變菌落數則大幅滑弱，這個結果就表示肝臟P-448形式參與MeIQx的活化。由Table 9、10、11亦可知Cytochrome P-450也是重要的活化酵素，在Cytochrome P-450中五種形式的酵素中以P-448-H具有最高的活化MeIQx的能力，其次為P-448 C，但是PCB誘發的酵素形式為P-450 b和P-450 e，因此僅能活化MeIQx很低的致突變性。在沒有化學物誘發的老鼠肝臟中P-448-H的活性也很低，但是雌鼠較雄鼠高兩倍，而所引發的反突變菌落數則雄的比雌的高，按照這樣的結果可能是P-450-male有較高的代謝活性，由Table 11可知P-450-male約佔整個肝臟中微粒體Cytochrome-P-450酵素活性的30～40%，因此在雄鼠中含量低亦能有效活化MeIQx和IQ的致突變性，這個結果也就反應了為何IQ化合物對不同性別的老鼠具有不同的致癌性的原因。

TABLE 8

Activation of MeIQx by liver microsomes from untreated, PB-, 3-MC- or PCB-treated rats

Treatment	Mutagenicity (rev/mg protein)	*N*-Hydroxylation (nmol/mg protein/10 min)
	Experiment 1	
Untreated male[a]	20 000 ± 2 100	0.15 ± 0.04
PB-treated male	32 000 ± 3 400	0.28 ± 0.02
3-MC-treated male	186 000 ± 23 000	1.73 ± 0.04
PCB-treated male	414 000 ± 65 000	3.00 ± 0.21
	Experiment 2	
Untreated		
Male	15 600 ± 1 450	
Female	7 860 ± 1 110	
PCB-treated		
Male	297 400 ± 21 200	
Female	289 400 ± 34 200	

Values given are mean ± SD.
[a]Six animals were used for each group.

Barnes和Weisburger（1985）[13]首先將IQ由尾靜脈注射入老鼠體中，收集其尿液測定致突變性，以了解IQ在體內代謝的途徑。如Fig. 11，顯示經過IQ注射的老鼠尿液對不同菌株的致突變性有不同的反應，對TA 98和TA 100均有線性濃度關係，而對TA 1537和TA1535則沒有反應。在Fig. 12則顯示不同濃度處理的老鼠的24小時尿液的致突變性亦有線性濃度關係。若以IQ 50mg／kg 體重的濃度給予老鼠，其尿液收集6小時、12小時、24小時和48小時，以研究IQ排出體外的時間如何？

結果如Fig. 13所示，間接致突變物（即IQ本身）較其代謝物排出的快，尿液中的間接致突變物，在24～48小時收的致突變性僅為0～6小時收的15%，而直接致突變物由尿液排出則在6～24小時為最高。直接致突變物僅佔間接

致突變物的0.3%。在整個48小時實驗過程收集到的致突變物的致突變性僅為原來給予的1%，其餘99%可能被身體吸收，經由其他排泄途徑排出體外，或是代謝為沒有致突變性的化合物排出體外。

TABLE 9

Effect of different inhibitors on the microsomal activation of MeIQx by hepatic microsomes from PCB- or 3-MC-treated rats

Inhibitor	Concentration (μM)	Mutagenic activity (rev/μg protein) PCB-treated		3-MC-treated	
None		348[a]	(100)[b]	106.0	(100)
Flavone	50	7	(2)	6.8	(6.3)
7,8-Benzoflavone	10	198	(56.8)	n.d.[c]	
	50	n.d.		25.2	(23.5)
Ellipticine	0.5	95	(27.3)	27.8	(26)
Metyrapone	200	172	(49.4)	68.0	(63.6)
SKF-525A	200	185	(53)	58.8	(55)

[a]The experiments were performed in duplicate and the mean value is presented. This experiment was repeated at least twice using hepatic microsomes obtained from different rats and no appreciable difference was observed between the two experiments.
[b]Numbers in parentheses are the relative activity.
[c]n.d., not determined.

TABLE 10

Requirement for reconstitution of the activating system

System	Mutagenic activity (rev × 10^{-3}/nmol P-450)	
Complete	684.5[a]	(100)[b]
Minus P-488-H	20.5	(2.9)
Minus reductase	10.2	(1.5)
Minus lipid	362.0	(52.8)

The complete system included P-488-H, NADPH-cytochrome P-450 reductase and lipid in the reaction mixture as shown in Materials and methods.
[a]Values are means of duplicate determinations.
[b]Numbers in parentheses are activities (%) relative to the complete system.

TABLE 11

Mutagenic activation of IQ and MeIQx by different forms of purified cytochrome P-450 preparations

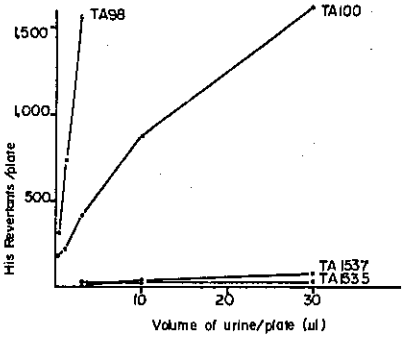
Form	Mutagenic activity (rev × 10^{-3}/nmol P-450) IQ		MeIQx	
P-448-H	1723[a]	(100)[b]	472	(100)
P-488-L	116.5	(6.7)	141	(29.8)
P-450b	19.7	(1.1)	7.4	(1.5)
P-450e	22.8	(1.3)	15.4	(3.3)
P-450-male	236	(13.6)	49	(10.4)

[a]Values presented are the means of duplicate determinations and consistent results were obtained in two separate experiments. The experimental details are described in Materials and methods.
[b]Numbers in parentheses are the activities (%) relative to the complete system.

由以上結果，我們可以確定IQ和其代謝物會經由尿液排出體外，而引起排泄途徑的器官表皮細胞癌化，由前面的結果也可知IQ會引起不按計劃的DNA合成，所以IQ會和哺乳類細胞DNA結合，這一點容於後面再詳加探討。

總之，肝臟是代謝IQ類化合物的主要器官，使它們轉變為有反應性的化合物和解毒的產物，例如肝臟Cytochrome-P-450酵素系統能活化IQ類化合物為N-hydroxylamino代謝物，然後進行O-sulfate和O-acyl結合反應形成最終致突變物和致癌物。Hayatsu等（1987）[14]以MeIQx餵老鼠，在尿和糞便中純化出它的代謝物，其化學構造經確定為2-amino-8-hydroxy-methyl-3-methylimidazo〔4,5-f〕quinoxaline，2-acetyl-amino-3,8-dimethylimidazo〔4,5-f〕quinoxaline和2-amino-8-methylimidazo〔4,5-f〕quinoxaline這些代謝物僅佔整個濃度的很小部份。最近Turesky（1988）[15]又由膽汁中確定五種主要代謝物，它們無論是否加有S_9都沒有致突變性，這五種代謝物經確定為如Fig. 14所示，其NMR的Chemical Shift如Table 12。作者在1988年除了確定五種MeIQx的膽汁代謝物外，並比較尿液和糞便中的代謝物。口服20mg／kg MeIQx的老鼠，在24小時內排出25～50%，而注射相同量的MeIQx的老鼠，在12小時內就排出40～75%。由HPLC分析尿液、糞便和膽汁中的MeIQx的代謝物，結果如Fig. 15所示。膽汁的主要代謝物爲1、3、5，尿液為1、4、5，而糞便則為6。各種MeIQx的代謝物，在尿液、糞便和膽汁中所佔的比例，如Table 13所示。由本實驗的結果又顯示大部份的MeIQx的代謝物，都不會成為致癌物來引起小腸和大腸癌。主要的MeIQx膽汁代謝物並不會從尿液排出體外，在放射性追踪實驗裏又無法由糞便中完全抽出，可能是在腸道中這三種主要代謝物會互相轉變而造成這樣的結果。

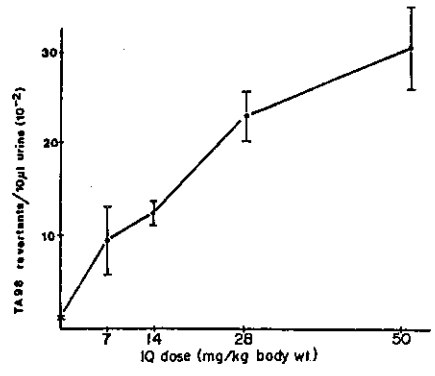
Fig. 11



Mutagenicity of urine from rats treated i.v. with IQ (50 mg/kg body wt.) in four Ames tester strains in the presence of S9 mix. Values are actual plate counts without subtraction of spontaneous revertants, that were 43±11 for TA98, 141±4 for TA100, 9±1 for TA1537, and 25±1 for TA1535 (see text).

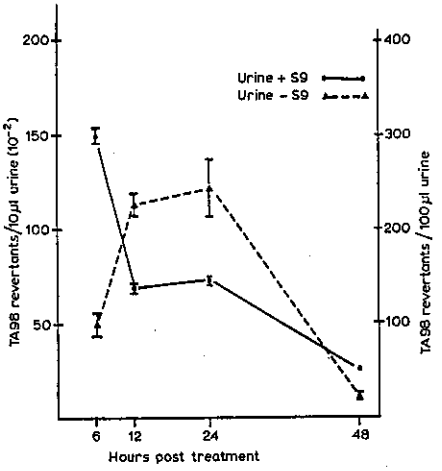
有許多實驗均證實異環狀胺類的致突變性是須要肝臟活化，同時老鼠的小腸微粒體也能活化它們。在無氧狀

Fig. 12



Dependence of urinary promutagen excretion on IQ dose administered.

Fig. 13

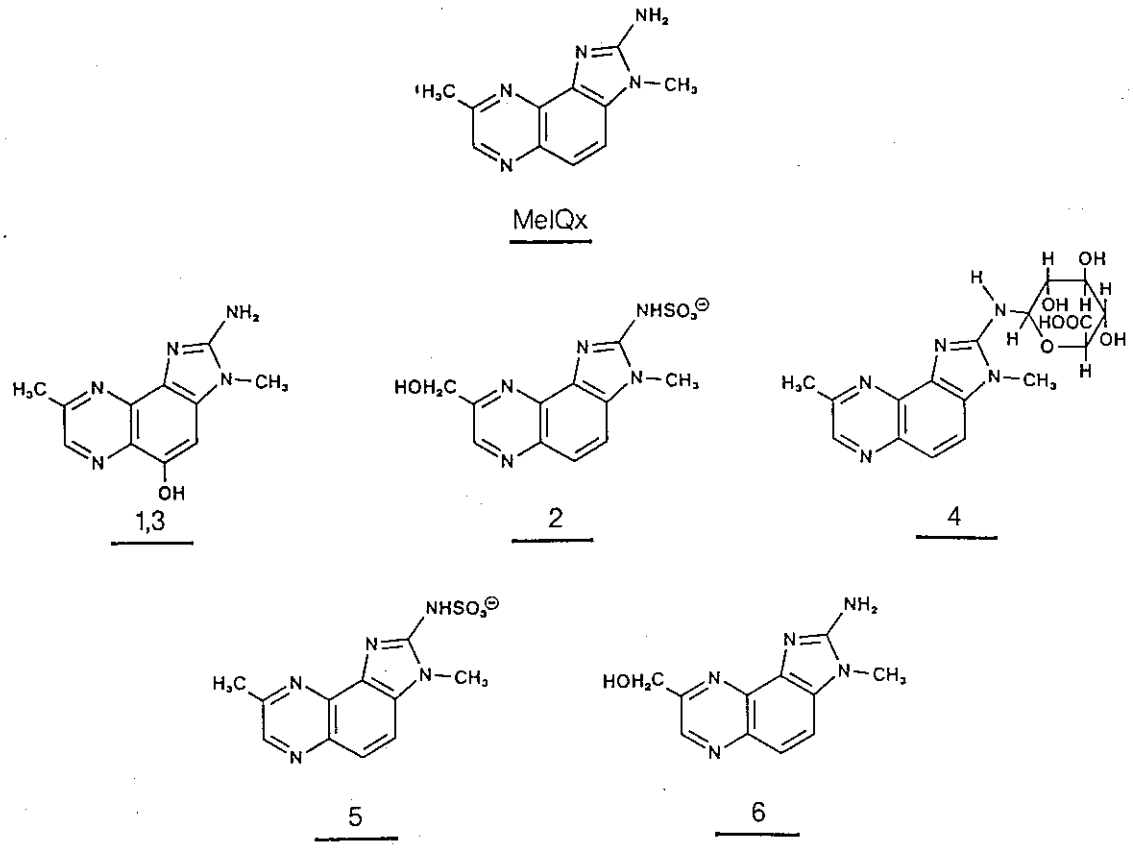


Kinetics of urinary mutagen excretion. Left ordinate refers to assays done with S9 mix. Right ordinate refers to assays without addition of S9.

態下，人類糞便微生物也能代謝IQ轉變為2-amino-3,6-dihydro-3-methyl-7H-〔4,5-f〕quinoline-7-one。MeIQx的膽汁代謝物，經由小腸mixed-function氧化酶作用會引起老鼠細胞的染色體變異。也有實驗證明小腸細胞中含有高量的前列腺素H合成酶亦能活化IQ成為致突變物。就如IQ一樣，MeIQx進行Sulfamate反應是主要的解毒過程，所以在糞便和尿液中，MeIQx Sulfamate是主要的代謝物。另一個結合反應，N-glucuronidation也是前驅致癌物解毒的主要途徑，其代謝物則由尿液排出體外。第1和3的MeIQx代謝物是在環的第五位置上羥化而形成，在致突變性上均無反應因而亦被解毒。由以此討論可知MeIQx的解毒作用和其他異環狀胺類化合物相同，大多進行環的羥化，Sulfation和glucuronidation，但是MeIQx不會形成N-hydroxylamine或它的hydroxamic acid。至於MeIQx在小腸微粒體酵素的代謝物仍須進一步的研究。

IQ化合物除了能被肝臟酵素系統活化之外，還能被人類腸道的微生物代謝成為直接的致突變物Carman（1988）[16]將這種經由Eubacterium菌種代謝的IQ代謝物由糞便中純化出來，其化學構造為2-amino-3,6-dihydro-3-methyl-7H-imidazo〔4,5-f〕quinoline-7-one（HOIQ），如Fig. 16所示。它的致突變性如Fig. 17已不須要S_9的活化，成為直接致突變物。IQ的衍生物經常因為有某一官能基或失去某一官能基而有不同的致突變性，例如去胺基或去甲烷基

Fig. 14



Proposed structures of metabolites of MeIQx. (1, 3) The primary metabolite which was conjugated with glucuronic acid (1) or acetylated (3), (2) *N*-(8-hydroxymethyl-3-methylimidazo[4,5-*f*]quinoxalin-2-yl)sulfamic acid, (4) N^2-(β-1-glucosiduronyl)-2-amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline, (5) *N*-(3,8-dimethylimidazo[4,5-*f*]quinoxalin-2-yl)sulfamic acid and (6) 2-amino-8-hydroxymethyl-3-methylimidazo[4,5-*f*]quinoxaline.

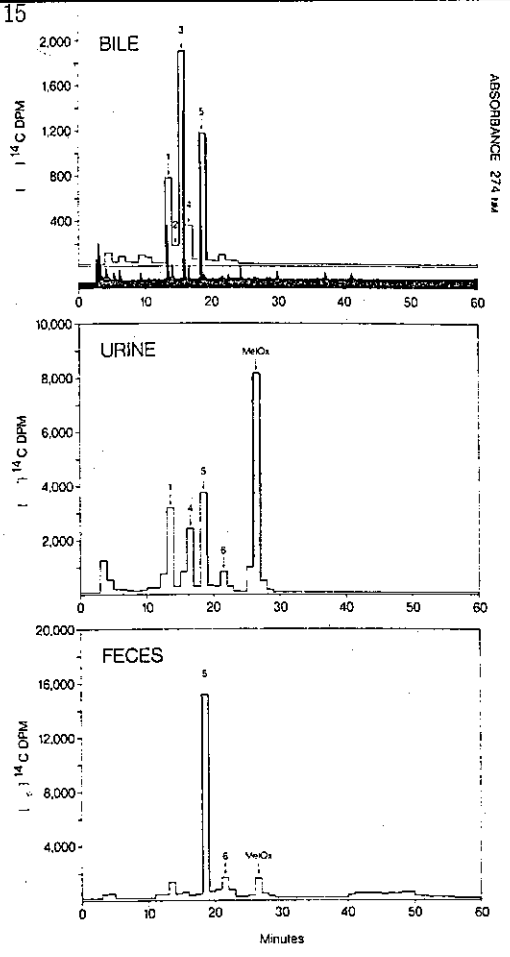
TABLE 12

500 MHz ^{1}H-NMR spectral parameters for the five metabolites of MeIQx isolated from bile (values are the chemical shift in p.p.m. from tetramethylsilane)

Assignment	Metabolite					MeIQx
	1	2	3	4	5	
8−CH_3	2.67	4.83[a]	2.77	2.70	2.63	2.73
N−CH_3	3.60	3.67	3.70	3.70	3.38	3.70
C_4−H	7.49	7.80[b]	7.94	7.60[b]	7.32[b]	7.75[b]
C_5−H	–	7.90[b]	–	7.75[b]	7.49[b]	7.87[b]
C_7−H	8.61	8.91	8.84	8.66	8.51	8.77
$COCH_3$	–	–	2.50	–	–	–

[a]In this metabolite the 8−CH_3 has been converted to a hydroxy methylene group which accounts for the large chemical shift relative to that of the other metabolites.
[b]Signals that appeared as doublets, J = 8.5 Hz.

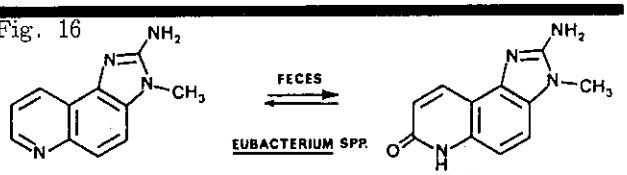
Fig. 15



HPLC analysis of metabolites isolated from bile, urine and feces from rats dosed intragastrically with [2-^{14}C]MeIQx (20 mg/kg body weight). A linear gradient containing 0.1 M ammonium acetate (pH 7.6) containing 5% methanol to 100% methanol over 60 min at a flow rate of 2 ml/min was used for purification. One-minute fractions were collected and measured for radioactivity.

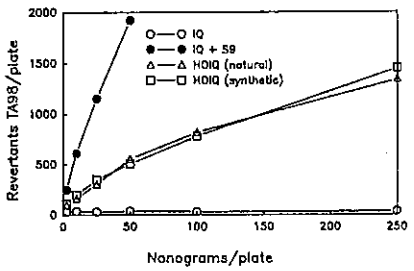
的IQ通常較IQ的致突變性為低。這些反應都可經由腸道中微生物的作用而發生，因此改變IQ了的遺傳毒性，但是很不幸的腸道微生物也代謝產生具有直接致突變性的HOIQ衍生物，而不須要經由肝臟微粒體酵素活化，直接作用於結腸的粘膜細胞造成腫瘤，實際上，在體內HOIQ如何形成？代謝途徑如何仍須要進一步探討。

Fig. 16



Metabolism of IQ to HOIQ by feces and pure cultures of Eubacterium spp.

Fig. 17



Mutagenicity of IQ and synthetic and natural (bacterially produced) HOIQ on TA98. Each value represents the average of duplicate plates at each dose in three separate assays. [spontaneous revertants/plate = 24–27].

IQ代謝物的DNA adduct

有關IQ代謝的研究最早是由日本學者Okamoto（1981）[17]開始，他確定IQ第一步代謝途逕是形成N-hydroxy-IQ，然後陸續有Yamazoe和Synderwine也都確認，合成N-OH-IQ（1983[18]，1987）[19]。Synderwine（1988）[20]又再進一步證明N-OH-IQ這個代謝物的直接致突變性有一部分會再代謝為N-acetoxy-IQ，是經由細菌的N-acetyl transferase的作用。同時他也發現N-OH-IQ在pH7.0時會和DNA共價結合，還能被細胞質內的O-acetyl transferase（哺乳細胞）進一步活化為N-acetoxy-IQ而增強與DNA的結合作用。化學致癌物和DNA的共價性相互作用是致癌機制的起始步驟中相當重要的關鍵步驟。雖然過去的許多研究都已知道DNA會與N-OH-IQ和N-acetoxy-IQ發生反應，但是它們的

TABLE 13 Disposition of metabolites of MeIQx (values are expressed as a percentage of administered dose determined by liquid scintillation counting of HPLC peaks)

	Metabolite						MeIQx
	1	2	3	4	5	6	
Urine	5.9 ± 1.8	–	–	5.2 ± 1.3	5.0 ± 3.6	2.0 ± 0.2	14.1 ± 10.5
Feces[a]	–	1.5 ± 0.5	0.9 ± 0.8	–	18.4 ± 0.2	2.0 ± 0.2	2.9 ± 0.3
Bile	3.8	0.6	10.8	0.6	5.4	–	0.3
	7.5	0.6	22.4	0.2	7.8	–	0.2

Three rats were dosed orally with MeIQx (20 mg/kg body weight) and urinary and fecal metabolite estimates were made after collection of excreta for 72 h. Bile estimates were obtained from two rats dosed orally at 20 mg/kg body weight and bile was collected for 24 h. Recovery of the dose in bile was 25 and 43% of the dose.
[a]Estimates of fecal metabolites are based on soluble radioactivity. The method of extraction recovered 39 ± 19% of the radioactivity in feces.

DNA adduct仍然沒有分離純化出來，Synderwine在1988年9月即用化學合成法合成出N-（deoxyguanosine-8yl）-IQ，並且以HPLC分離出N-OH-IQ和N-acetoxy-IQ的主要DNA adduct。Table 14的結果顯示每一毫克核酸和N-OH-IQ的反應能力，依序為poly G＝DNA≫poly A＞poly C＝poly U。若加入醋酸酐則其反應能力提高6.4和2倍。大約有6%的N-OH-IQ會與DNA作用（在醋酸酐存在時），反之則僅有1%會與DNA發生反應。但是N-OH-IQ與poly C和poly U的結合反應不會因加入醋酸酐而有所改變。在另一個實驗作者用〔^{3}H〕-OH-IQ和deoxyguanosine和醋酸酐一起發生反應，並以HPLC分離出具有放射性的反應物在retention time36～37分鐘出現如Fig.18所示。有一個未知的代合物在19－20分鐘出現，它不具有放射性，可能是deoxyguanosine的acetylated衍生物。t^{R}36～37的放射性反應物經UV、MS、NMR的分析如Fig. 2、3所示UV λ max為250.4、293.5和342.5nm，分子量為463.4，由NMR圖可看到N-Methyl group和五個芳香環的質子Chemical shift，這表示有替代反應發生在IQ的胺基氮原子上。另外五個去氧核醣碳上的質子也能在圖譜上看到（H-1′、3′、4′、5′、5′）。尤其是H^8-guanyl resonance大約在～8.0 ppm處已消失掉表示IQ被此嘌呤的碳原子結合，形成N-（deoxyguanosine-8-yl）-IQ，這也就是IQ的代謝物N-OH-IQ和N-Acetoxy-IQ在體外與DNA結合形成的DNA adduct，至於在體內是否亦會形成則須要再加以研究。（Fig. 19、20）

最近Negishi（1989）[21]實驗發現MeIQx會和小牛胸腺DNA和小牛肝臟DNA結合。這個經肝臟酵素活化代謝的MeIQx代謝物也被HPLC純化分離確定是2-hydroxy-amino-3.8-dimethylimidazo〔4,5-f〕quinoxaline（N-OH-MeIQx）。後來也就以人工合成的N-OH-MeIQx進行一系列的實驗。即使在pH中性，0℃狀態，添加醋酸酐都能促進N-OH-MeIQx和DNA的結合增加10倍。這個結果告訴我們N-OH-MeIQx是MeIQx經由微粒體酵素P-450代謝形成，然後再進

TABLE 14

Binding of [^{3}H]*N*-hydroxy-IQ to DNA and polynucleotides in the presence and absence of acetic anhydride[a]

	Acetic anhydride	pmol bound/mg[b]
Polyguanylic acid	–	70.3 ± 10.8
	+	274.7 ± 9.1
Polyadenylic acid	–	16.3 ± 2.3
	+	28.1 ± 2.1
Polycytidylic acid	–	10.5 ± 0.1
	+	9.7 ± 0.9
Polyuridylic acid	–	8.3 ± 1.2
	+	8.6 ± 0.5
DNA (pH 7.0)	–	58.8 ± 2.4
	+	334.1 ± 21.5
DNA (pH 5.0)	–	65.7 ± 2.0

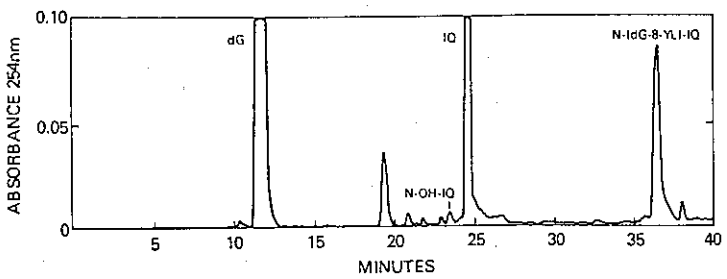
[a]As described in Materials and methods, the reaction mixture contained DNA or polynucleotides in 70 mM Sorenson's phosphate buffer, pH 7.0, [^{3}H]*N*-hydroxy-IQ and, where indicated, acetic anhydride. [^{3}H]*N*-Hydroxy-IQ was also reacted with DNA in 70 mM Sorenson's phosphate buffer, pH 5.0.
[b]Values are the mean ± standard error of two experiments run in duplicate

一步乙醯化（Acetylated）而和肝臟DNA結合。由Table 15可知，在沒有醋酸酐存在的狀態下N-OH-MeIQx對小牛胸腺DNA和加熱過的小牛胸腺DNA的結合力沒有差異，但是在醋酸酐存在時，單鏈DNA較不然小牛胸腺DNA的結合力高4倍，當N-OH-MeIQx和2′-deoxyguanine反應會形成DNA adduct。

N-OH-MeIQx在沒有S$_9$活化下就能引起TA98 150000 rev／μg，對TA100則為960rev／μg，如Fig. 21、22所示。它的致突變性能被PCP抑制，但是不能被DCNP抑制，若加入S$_9$的Cytosolic fraction則能促進N-OH-MeIQx六倍的致突變性。

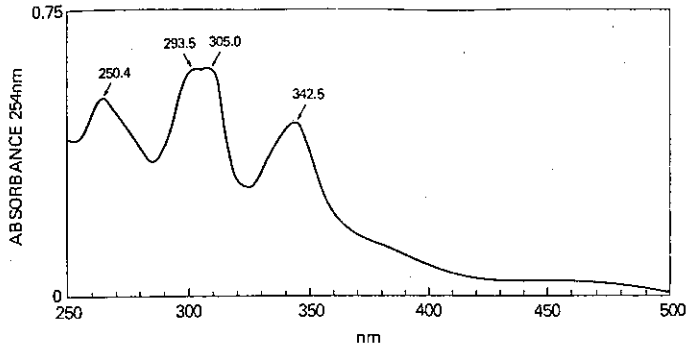
總之，N-OH-MeIQx是經由肝臟酵素P-448活化後，和DNA結合而造成致突變性，它解pH7.0時發生DNA adduct，而N-OH-trp-p-2則在pH5.0，而N-OH-Glu-P-1則必須有進一步酯化才能與DNA結合。加入醋酸酐的目的是在增強N-OH-MeIQx和DNA的結合力。而MeIQx的乙醯化代謝物N-acetyl-MeIQx則須要代謝活化後才會有致突變性，它是經由O-acetylation將N-OH-MeIQx轉變為高

Fig: 18



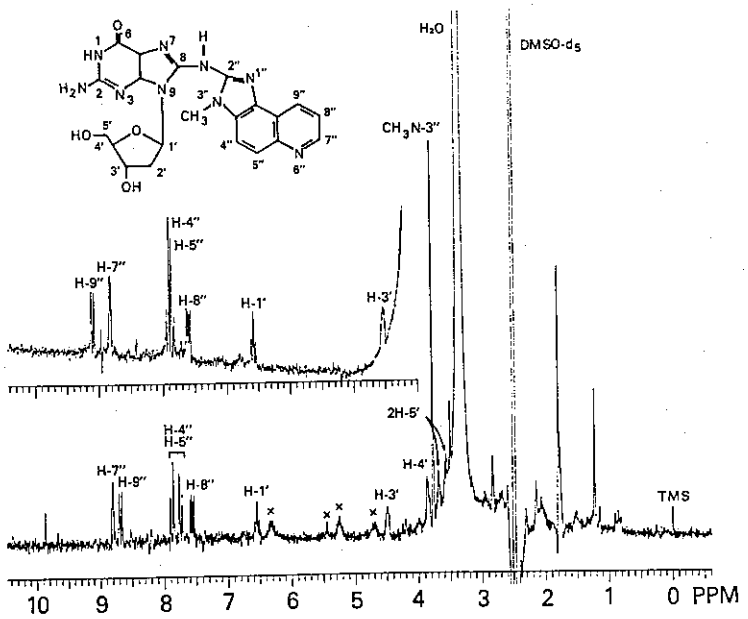
HPLC profile of crude synthetic *N*-(deoxyguanosin-8-yl)-IQ. The reaction mixture for *N*-(deoxyguanosin-8-yl)-IQ synthesis was analyzed on a Supelcosil C_{18} analytical column (LC-18-S, 25 cm × 4.6 mm, 5-μm particle size) after partial purification with a C18 Sep-Pack. The solvent system was methanol/aqueous 0.05 M KH_2PO_4 (degassed) set at a flow rate of 2 ml/min. Solvent programming was a linear methanol gradient from 1 to 4% by 5 min, to 12% by 10 min, isocratic from 10 to 15 min, from 12% to 50% by 30 min, and to 100% by 40 min post-injection.

Fig. 19



UV–visible spectrum of HPLC-pure *N*-(deoxyguanosin-8-yl)-IQ in methanol/water (50:50).

Fig. 20



200-MHz proton NMR spectrum of *N*-(deoxyguanosin-8-yl)-IQ. ~50 μg in 0.5 ml DMSO-d_6. Chemical shift values = δ 8.80 (dd, H-7"), 8.68 (brd, H-9"), 7.88 (d, H-4"), 7.74 (brd, H-5"), 7.57 (dd, H-8"), 6.55 (t, H-1"), 4.50 (br, H-3"), 3.86 (br, H-4"), 3.70 and 3.60 (br, 2H-5"), 3.77 p.p.m. (s, CH_3N-3"). Signals marked (×) are exchangeable with D_2O and correspond to the NH and OH protons. The two H-2' protons are obscured by the large solvent signal in the 2.0–2.8 p.p.m. region. Coupling constants: $J_{4,5}$ = 9.0 Hz, $J_{7,8}$ = 4.3 Hz, $J_{7,9}$ = 1.7 Hz, $J_{8,9}$ = 8.3 Hz, $J_{5,9}$ = 0.6 Hz. Inset: NMR of adduct in 0.5 ml DMSO-d_6 and 50 μl D_2O. Note: in addition to exchange of protons on heteroatoms, there is apparently a solvent effect on the chemical shift of the H-9" proton. The small sharp proton-exchangeable signal at 9.85 p.p.m. was not assigned.

親電子性的2-N-acetoxyamino-3.8-dimethyl-imidazo〔4,5-f〕quinoxaline（N-Acetoxy-MeIQx），在老鼠肝臟Cytosol fraction含有許多酵素能催化acetylation，例如arylamine N-acetyl transferase、N-hydroxy arylanine、O-acetyltransferase 和arylhychoxyamic acid N,O-acyl-transferase，因此N-OH-MeIQx能進行Acetylation後與肝細胞DNA結合。另外Sulfotransferase和aminoacyl-t-RNA synthetase能進一步活化N-hydroxylated amine，例如2-acetylaminofluorene、4-aminoquinoline和Trp-P-2。

N-OH-MeIQx也能在醋酸酐存在或不存在狀態下和poly G結合，然後用DNase-I和alkaline phosphatase分解，其水解物以HPLC分離。它的主要成份為2'-deoxyguanose adduct，由此結果可知老鼠肝細胞DNA的guanine是主要被修飾的鹼基。同樣的結果也發現在Trp-P-2和Glu-P-1，也形成C-8的guanine adduct。

N-acetoxy-Glu-P-1是Glu-P-1的可能最後代謝物能有效的與天然DNA反應。尤其是在G-C Cluster的區域，例如在PBR322 DNA的這些區域發生反應，這也就表示N-acetoxy-Glu-P-1能插入（intercalation）到DNA中，而N-OH-MeIQx就有剛好相反的特性，它比較喜歡和單鏈poly G DNA作用，同時和poly（G-C）的結合力也較天然小牛胸腺DNA為高，可是在修飾DNA的作用上，則沒有差異，這表示N-OH-MeIQx和它的acetylated形式插入DNA的作用上，與致突變的形成無關。從另一方面而言，IQ型化合物和DNA的混合液的UV吸光光譜也顯示這種插入的胺

類是非共價鏈性的。由Negishi（1989）的實驗較傾向於認為N-OH-MeIQx與DNA形成adduct較intercalation的機會為大。

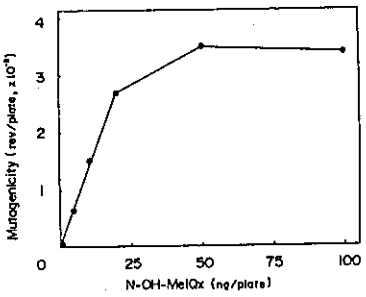
由PCP能抑制MeIQx的致突變性的結果，可知N-OH-MeIQx須要進一步被O-acetyltransferase活化，最近由TA98分離純化出此種酵素，它的活性能被PCP抑制，所以這個酵素在MeIQx的代謝活化上非常重要。但是在Trp-P-2則不能被PCP抑制。我們知道N-hydroxylation是異環狀胺類化合物共同的活化途徑，可是或許它們有不同的esterification步驟，而使得它們對PCP有不同的反應。

TABLE 15

BINDING OF N-OH-MeIQx TO HOMOPOLYMERS OR RIBONUCLEIC ACID IN THE PRESENCE OR ABSENCE OF ACETIC ANHYDRIDE

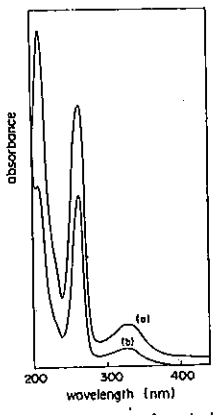
	Without acetic anhydride (nmole/mg polymer)	With acetic anhydride (nmole/mg polymer)
poly G	3.3 ±0.22	25 ±0.45
poly C	0.07±0.02	0.48±0.07
poly A	0.02±0.01	0.24±0.01
poly U	0.02±0.01	0.24±0.03

Fig. 21



Mutagenicity of N-OH-MeIQx to *Salmonella typhimurium* TA98.

Fig. 22



UV spectra of synthetic N-OH-MeIQx (a) and peak 1 (b) in 40 mM potassium phosphate buffer, pH 2.

結語

由以上的討論，IQ化合物的哺乳細胞遺傳毒性和代謝途徑，代謝化合物以及它們的DNA adduct有下列數點結論。

1.IQ類化合物對細菌能引起點突變，故在肝臟酵素活化是一很強的致突變物，可是對哺乳細胞的遺傳毒性則由於哺乳細胞的解毒酵素活性甚高，再加上DNA修補系統超強故對哺乳細胞僅為弱的遺傳毒物。

2.IQ在體內代謝後僅有1%的致突變性由尿液排出體外，其餘99%則可能被體內代謝為沒有致突變性的化合物或經由其他排泄途徑送出體外。主要參與代謝的器官為肝臟，在肝細胞含有Cytochrome-P-450酵素系統能活化代謝IQ化合物，首先經由N-hydroxylation，然後Acetylation，最後進行Conjugation，具有致突變性成致癌性的化合物為N-OH-IQ或N-OH-MeIQx以及N-Acetoxy-MeIQx等，而再經由O-Sulfate和O-acyl Conjugation後則變為沒有致突變性的代謝物，例如MeIQx則有五種膽汁代謝物。

3.IQ除了能被肝臟酵素系統代謝外，還能被人類腸道中的微生物代謝，例如Eubacterium spp.可將IQ轉變為HOIQ，它是一種直接致突變物，因此有可能成為大腸癌的起始劑。

4.Synderwine（1988）首先分離純化確定N-OH-IQ和N-Acetoxy-IQ的DNA adduct為N-（deoxyguanosine-8-yl）-IQ，Negishi（1989）也將MeIQx的DNA adduct純化確定為2′-deoxyguanosine adduct，因此IQ代謝物主要與DNA的2′-deoxyguanosine發生鍵結形成DNA adduct進而引起癌化的起始步驟。

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