SYNTHESIS OF FLUORINE ANALOGS OF NATURAL PORPHYRINS POTENTIALLY USEFUL FOR DIAGNOSIS AND THERAPY OF CANCER

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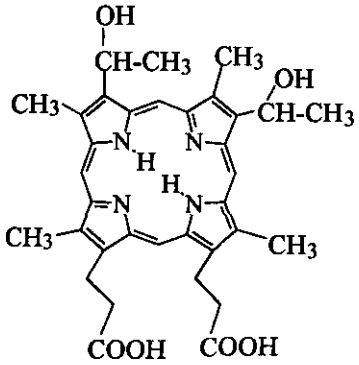
This is dedicated to the memory of the late Professor Yoshio Ban.

Abstract----Hematoporphyrin derivative (HpD) or Photofrin II are used for photosensitizer of photodynamic therapy (PDT) of cancer. However, these are complex mixtures of porphyrin derivatives. We have synthesized fluorine analogs of naturally important porphyrin derivatives, such as protoporphyrin and hematoporphyrin, which would be useful for diagnosis and therapy of cancer. In this review, we wish to show our syntheses of these fluorine analogs and the localization of these porphyrins to tumor cells and some tissues.

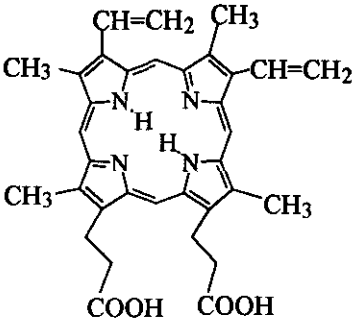
INTRODUCTION

Porphyrin is a planar macrocycle which consists of four pyrrole rings joined by four methine bridges. This macrocycle is highly conjugated and deeply colored. The main absorption bands have very high extinction coefficients, and the intense 'Soret' band, found around 400 nm, is a characteristic of this macrocyclic conjugation.[1] Furthermore, they show a characteristic red fluorescence by irradiation with a ultraviolet light. So, these porphyrin derivatives are investigated for application to various fields using their structural and spectroscopic properties. One of the recent investigations in these fields is the application of hematoporphyrin derivatives (HpD) to diagnosis and therapy of tumors.[2] It is well known that some porphyrin derivatives localize to tumor tissues,[3] and especially hematoporphyrin derivatives (HpD), obtained by treatment of hematoporphyrin (**1**) with sulfuric acid and acetic acid, is reported to localize to tumor tissues easily. After administration of HpD, a tumor tissue containing HpD fluoresces a reddish color by photoirradiation with a laser light, so the early stage of a cancer could be detected. Furthermore, it is known that some porphyrins

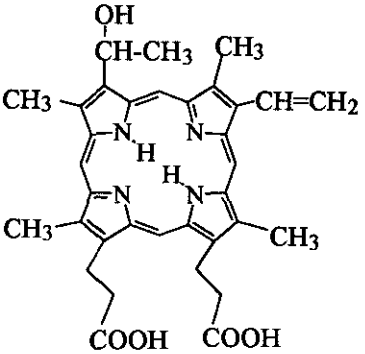
produce active oxygen by photosensitization and then destruct the cancer cells. This therapy of cancers by irradiation is called photodynamic therapy (PDT). However, HpD used for PDT is a complex mixture of several porphyrins, such as hematoporphyrin (HP: **1**), hydroxyethylvinyldeuteroporphyrin (HVD: **3**), protoporphyrin (PP: **2**), hematoporphyrin diacetate (HDA), and so on.[4]



Hematoporphyrin **1**



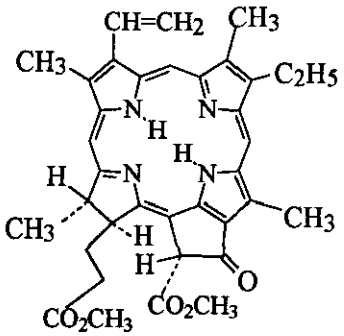
Protoporphyrin **2**



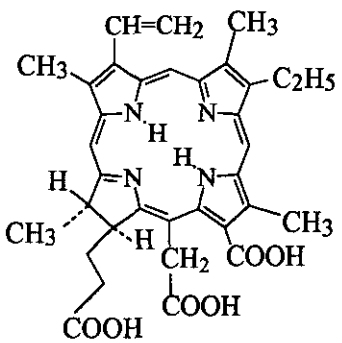
Hydroxyethylvinyl-deuteroporphyrin **3**

Figure 1

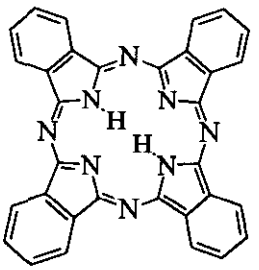
Therefore, HpD has some difficulties in clinical use; namely its low purity and unstable photosensitivity. Recently, Photofrin II was developed as a low photosensitivity drug. However, it is thought that this drug consists of dihematoporphyrin ester or ether as a main component. Actually, the composition is not constant. It is ambiguous which of the ester or ether is effective, because Photofrin II is not of constant composition as medicine and not pure.[5] Further, it is not clear what component of the mixture localizes to cancer cells. Therefore, on searching more effective photosensitizer than Photofrin II, syntheses and applications of phthalocyanines, chlorins,[6] hematoporphyrin oligomers (HPO),[7] Ga-complexes of porphyrin dyes,[8] and pheophorbide derivative (PH-1126)[9] are being investigated actively.



Pheophorbide-a



Chlorin-e_6



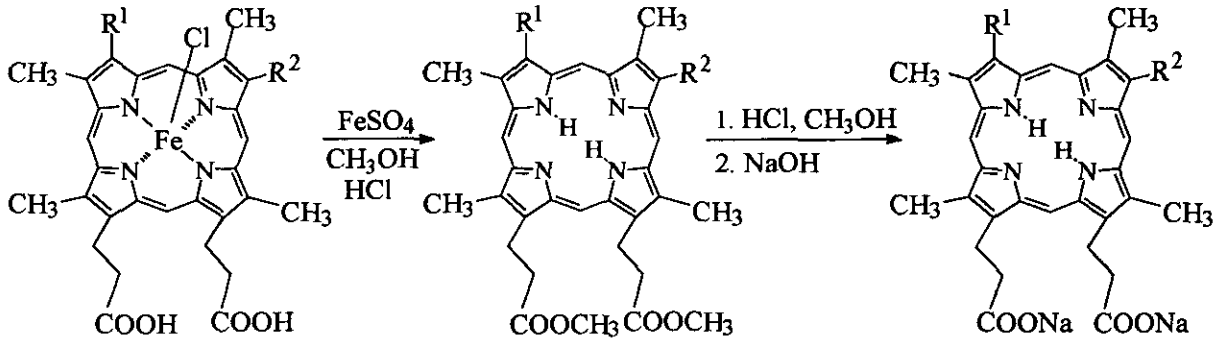
Phthalocyanine

Figure 2

Photosensitizer (the ground state of photosensitizer: S_0) is excited by absorption of photoenergy on irradiation. The excited singlet state ($^1S^*$) of this molecule changes to excited triplet state ($^3S^*$) *via* intersystem crossing (ISC). The energy of this excited triplet state is transferred to ground state oxygen ($^3O_2$) and excited singlet oxygen ($^1O_2^*$) is produced. The excited singlet oxygen is highly oxidative and reactive compared with ground state oxygen, so it is called as "active" oxygen. Another active oxygens produced are superoxide, hydroxyl radical, and hydrogen peroxide. These active oxygens are produced in cancer tissues by photosensitization of localized porphyrins, and oxidize important parts of cancer cells. As a result, cancer cells are deactivated and then cancer tissue is necrotized. In the photosensitized reaction by using porphyrin derivatives,[10] it is believed that excited singlet oxygen is formed at primary stage. However, contribution of radical types of active oxygens cannot be eliminated.[11] A photosensitizer itself returns to the ground state after transfer of the excited triplet state energy to ground state oxygen. In successive turns, it is excited by further irradiation, and these steps are repeated until the photoirradiation is stopped.

As mentioned above, Photofrin II used at present for PDT is a complex mixture[5] and not pure, so that this will not give constant results on localization to cancer and photosensitivity. Thus, if a pure porphyrin derivative, which has a high selectivity for a tumor tissue and is localized to special tumor cells, is discovered, it will be very useful for the diagnosis and therapy of cancers, and PDT will be developed greatly to allow the specific therapy against each cancer.

Previously, we reported[12] that 3-(1-methoxyethyl)-8-vinyldeuteroporphyrin sodium salt (**6**), 8-(1-methoxyethyl)-3-vinyldeuteroporphyrin sodium salt (**7**) and 3,8-bis(1-methoxyethyl)deuteroporphyrin sodium salt (**8**) were obtained by treatment of protoporphyrin dimethyl ester (**5**) with HCl gas in CH_3OH, followed by alkaline hydrolysis of the three CH_3OH-adducts of protoporphyrin, as shown in Scheme 1.



Hemin **4**

R^1, R^2 : -CH=CH$_2$

***PPDME* 5**

R^1, R^2 : -CH=CH$_2$

	R^1	R^2
6:	-CH(OCH$_3$)CH$_3$	-CH=CH$_2$
7:	-CH=CH$_2$	-CH(OCH$_3$)CH$_3$
8:	-CH(OCH$_3$)CH$_3$	-CH(OCH$_3$)CH$_3$

Scheme 1

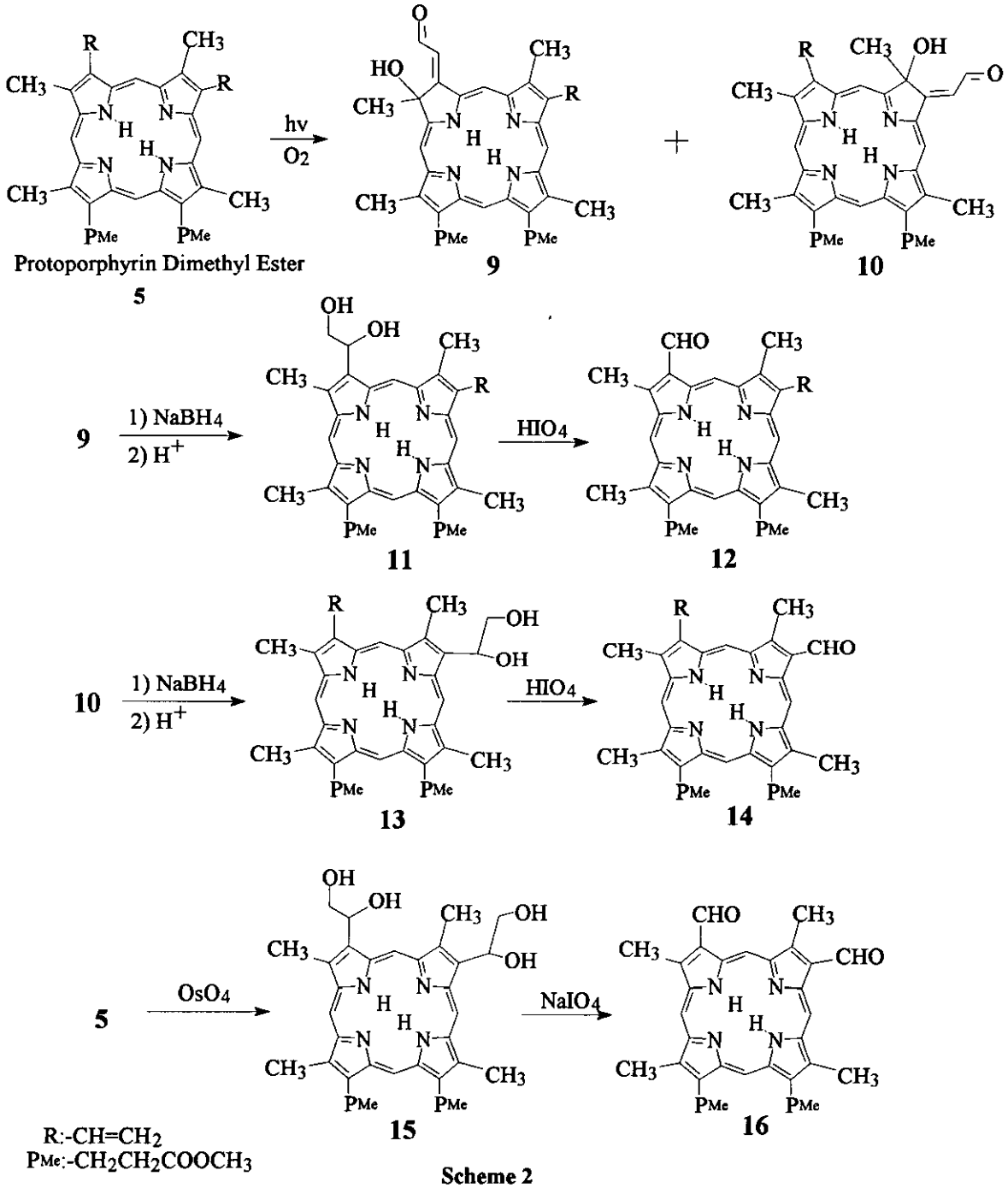
Investigation of the localization of each CH_3OH-adduct to human gastric cancer (GCA-1) showed that **6** had an interesting physiological property. Thus, **6** localizes specifically to gastric cancer, but does not localize to human hepatocellular carcinoma (HCC-1), while **7** and **8** localize little to both cancer cells. The compound (**6**) showed a similar photodynamic effect to JTC-16 cells on irradiation of laser as HpD.[13] These results suggest that some porphyrins might localize selectively to a specific tumor tissue and could be a specific sensitizer for a special kind of tumor. Since this work, we have been investigating synthesis of fluorine analogs of porphyrin derivatives which will localize selectively to a specific tumor tissue and a potentially useful for diagnosis and therapy of cancer. The reason why we chose fluorine analogs are: 1. A fluorine atom is as small as a hydrogen atom, and the incorporation of a fluorine does not change the shape of the original porphyrin, and will be taken up as the unsubstituted one. 2. Usually, fluorine compounds are very stable, but some are very reactive and these reactivities are believed to show important biological effects.[14] Some of the fluorine analogs were expected to react with biological components and to show anti-tumor effects. 3. Fluorine compounds are quite rare in biological systems. Therefore, if a fluorine analog localized to tumor tissues, it would be detected by F-nmr imaging. Now, we review the synthesis of fluorine analogs of natural porphyrin and preliminary biological test.

1: SYNTHESIS OF FLUORINE DERIVATIVES OF PROTOPORPHYRIN

Protoporphyrin (**2**) is a demetalated porphyrin of protoheme which is a component of hemoglobin. The zinc derivative is used as a medicine. Thus, **2** is also important as a bioactive compound, and we tried the synthesis of fluorine analogs of **2**. As mentioned above,[13] activity of CH_3OH-adduct of **2** depends on the site of addition, so that we thought that the vinyl groups of protoporphyrin play an important role. Therefore, first we investigated introduction of fluorine atom to the vinyl groups.

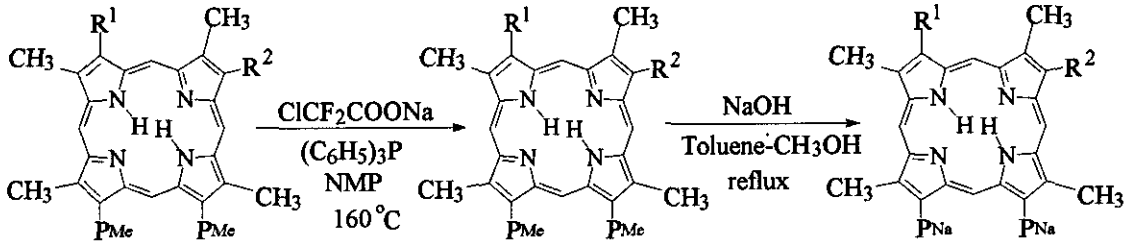
Protoporphyrin dimethyl ester (**5**) obtained from protohemin (**4**) was photooxidized by white light in the presence of oxygen to give so-called photoporphyrins (**9** and **10**), both isomers of which were separated by column chromatography. These compounds were converted to 3-formyl-8-vinyl- and 8-formyl-3-vinyl-deuteroporphyrin (**12** and **14**) by reduction of **9** and **10**, followed by rearrangement of the diols to glycol (**11** and **13**) and cleavage of the glycols (**11** and **13**) according to the literature[15] with some modifications,[16] since the yields of these formyl derivative (**12** and **14**) were much lower than reported. Thus, the reduction of photoporphyrins (**9** and **10**) with $NaBH_4$ was carried out at room temperature[16] instead of heating on a steam bath,[15] since formation of tarry substances was fairly large at a high temperature. After acidification and extraction with CH_2Cl_2, the glycols (**11** and **13**) were purified by column chromatography in 80 % yields. These glycols were oxidized in benzene-CH_2Cl_2 with $NaIO_4$-H_2SO_4 according to the literature,[15] but the yields of the desired formyl derivatives (**12** and **14**) were much lower than that shown in the literature. We thought that the

solubility of glycols (**11** and **13**) to this solvent system is very low, and this reaction occurred under a heterogeneous condition. Then, we tried the oxidation of the glycols in dioxane with HIO_4-$2H_2O$, and obtained the desired formyl derivatives in 80 % yields. Bis-formyl derivative (**16**) was synthesized by OsO_4-oxidation of protoporphyrin dimethyl ester (**5**), and subsequent $NaIO_4$-oxidation.[17] These are shown in Scheme 2.



Scheme 2

We tried difluorovinylation of these formyl derivatives (**12**, **14** and **16**) with sodium chlorodifluoroacetate in the presence of triphenylphosphine. Thus, reaction of the formyl derivatives (**12**, **14** and **16**) with sodium chlorodifluoroacetate and triphenylphosphine in *N*-methylpyrrolidone (NMP) gave difluoro- and tetrafluoro-protoporphyrin derivatives (**17**, **18** and **19**). Use of NMP as a solvent is essential. Diglyme, which is commonly used for this type of synthesis, did not give a good result (see Scheme 3.)



	R^1	R^2
12:	-CHO	-CH=CH$_2$
14:	-CH=CH$_2$	-CHO
16:	-CHO	-CHO

	R^1	R^2
17:	-CH=CF$_2$	-CH=CH$_2$
18:	-CH=CH$_2$	-CH=CF$_2$
19:	-CH=CF$_2$	-CH=CF$_2$

	R^1	R^2
3-FPPN (20):	-CH=CF$_2$	-CH=CH$_2$
8-FPPN (21):	-CH=CH$_2$	-CH=CF$_2$
3,8-FPPN (22):	-CH=CF$_2$	-CH=CF$_2$

P$_{Na}$:-CH$_2$CH$_2$COONa

Scheme 3

These difluorovinyl derivatives (**17**, **18** and **19**) were hydrolyzed with NaOH in toluene-CH_3OH to give sodium salts (**20**, **21** and **22**), as shown in Scheme 3, and the each sodium salt was subjected to the preliminary test of uptake by human gastric cancer (MKN-45). The results are shown in Figure 3. This figure shows that 8-FPPN (**21**) has a high localizability to cancer cells and stomach, and localized specifically to gastric cancer cells. When these were given to rat ascite hepatoma cells, 3,8-FPPN (**22**) was taken up most efficiently.[16] These facts suggested that a special cancer takes up a special porphyrin.

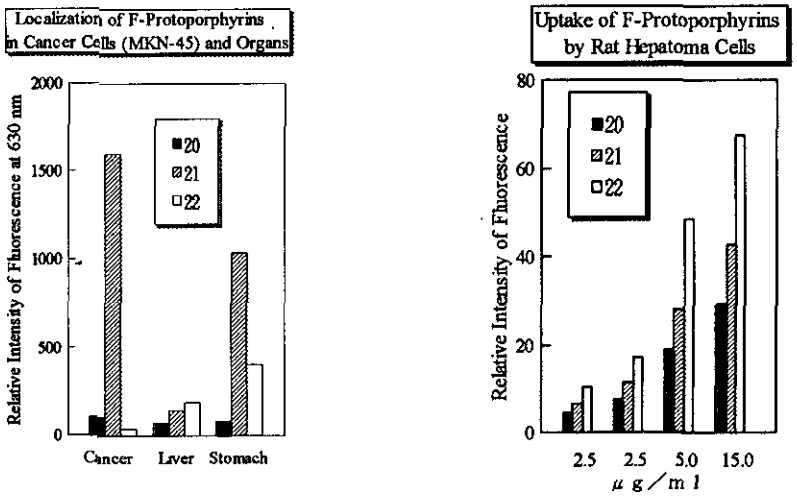
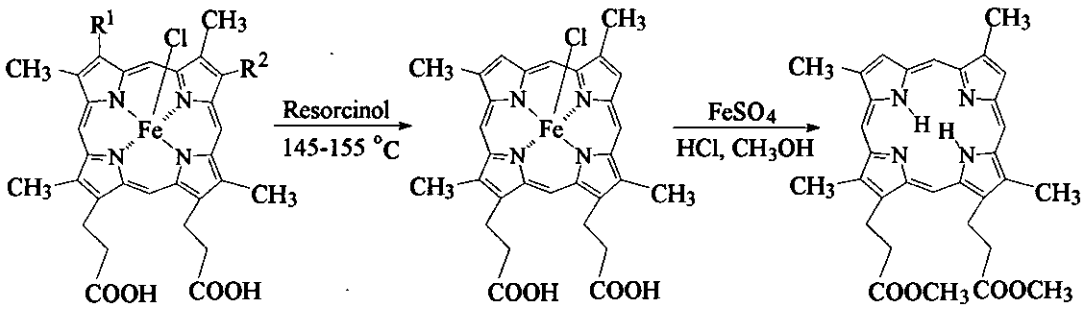


Figure 3

These results support that one porphyrin is taken up specifically by one cancer and other porphyrin by other cancer. Thus, we planned to synthesize fluorine analogs of other natural porphyrin derivatives and to investigate their localization to cancers.

2. SYNTHESIS OF FLUORINE ANALOGS OF HEMATOPORPHYRIN

Hematoporphyrin derivative (HpD), which was used as a photosensitizer of PDT, was derived from hematoporphyrin (**1**) by treatment with sulfuric acid and acetic acid, but this is a complex mixture of several porphyrins. So, we tried synthesis of pure fluorine analog of hematoporphyrin. By heating with resorcinol at 160 °C,[18] hemin (**4**) are converted into deuterohemin (**23**), and subsequent demetalation with $FeSO_4$-HCl in CH_3OH-$CHCl_3$-pyridine and esterification of deuterohemin (**23**) gave deuteroporphyrin dimethyl ester (DPDME: **24**) in 68 % yield (see Scheme 4). We tried to introduce fluorine substituent to **24**.



Hemin **4** R^1, R^2 : -CH=CH_2 **23** **DPDME 24**

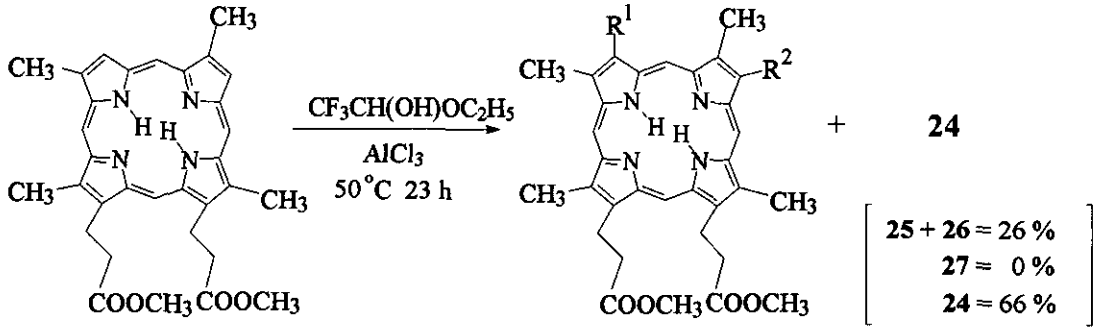
Scheme 4

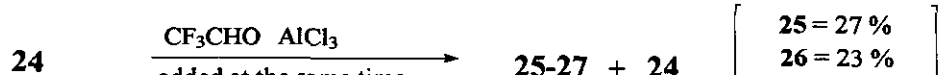
Guy *et al.*[19] reported synthesis of trifluorohydroxyethyl (TFHE) benzene derivatives by the reaction of benzene with trifluoroacetaldehyde ethyl hemiacetal in the presence of a Lewis acid. So, we planned to synthesize TFHE-containing DPDME using Lewis acid catalysts. Several Lewis acids ($FeCl_3$, $TiCl_4$, $SnCl_4$, $SbCl_5$, $ZnCl_2$ and $AlCl_3$) were examined as a catalyst, but the yields of TFHE-DPDMEs (**25** and **26**) were only 26% yield in total, and the starting material (**24**) was recovered in 66 % yield, when $AlCl_3$ was used. Other Lewis acids were less effective (see the top of Scheme 5.)

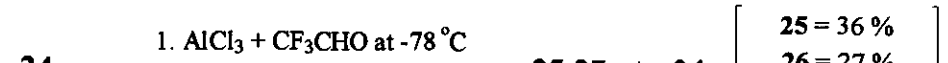
Then, we tried to introduce TFHE group to **24** by Friedel-Crafts reaction using CF_3CHO itself to increase the reactivity. Reaction of **24** with CF_3CHO in the presence of $AlCl_3$ gave mono-TFHE compounds (**25** and **26**) and bis-TFHE one (**27**), in 45-50% and few % yield, respectively. Thus, the yields were improved considerably (see the middle of Scheme 5.)

Further, after formation of $AlCl_3$ - CF_3CHO complex in CH_2Cl_2 at -78 °C, **24** was added to the solution of the complex, and reacted at 50 °C for 4 h. By this improvement of the procedure, mono-TFHE compounds (**25**

and **26**) were obtained in 36 and 26 %, respectively, and bis-TFHE (**27**) compound was obtained in 6 % yield with recovery of the starting material (**24**) (13%) (see the bottom of Scheme 5.)



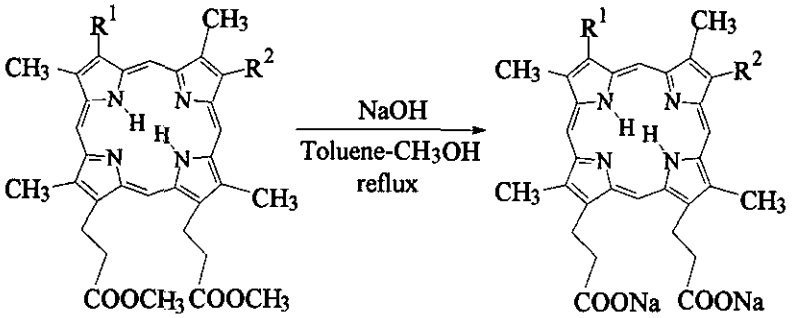




	R^1	R^2
3-TFHE-DPDME (25):	$CH(OH)CF_3$	H
8-TFHE-DPDME (26):	H	$CH(OH)CF_3$
3,8-TFHE-DPDME (27):	$CH(OH)CF_3$	$CH(OH)CF_3$

Scheme 5

These porphyrin derivatives (**25** - **27**) were hydrolyzed to give Na salts (**28** - **30**) (see Scheme 6). Then, uptake



	R^1	R^2
3-TFHE-DPN (28):	$CH(OH)CF_3$	H
8-TFHE-DPN (29):	H	$CH(OH)CF_3$
3,8-TFHE-DPN (30):	$CH(OH)CF_3$	$CH(OH)CF_3$

Scheme 6

of the Na salts (**28 - 30**) to human liver cancer cells (JTC-16) was investigated. Each TFHE-DPN (**28 - 30**) was added to the culture media of JTC-16 cells and incubated for 48 h. Then, the cells were washed with buffer solution and extracted with $(iso\text{-}C_3H_7)_2NH$-CH_3OH.[20] The intensity of fluorescence of this extract was measured by fluorophotometry. The results are shown in Figure 4. From these results, 3,8-TFHE-DPN (**30**) was found to be taken up more effectively to human liver cancer cells than other TFHE-DPN (**28** and **29**). Furthermore, of mono-TFHE compounds (**28** and **29**), the 8-TFHE derivative (**29**) was taken up more effectively to the cancer cells. It is speculated that the difference of these uptakes is concerned with increase of lipophilicity due to the fluorine substituents, and if the mechanism of uptake of porphyrins to cells is clarified, it will be possible to design porphyrin derivatives that will localize selectively to special tumor cells.

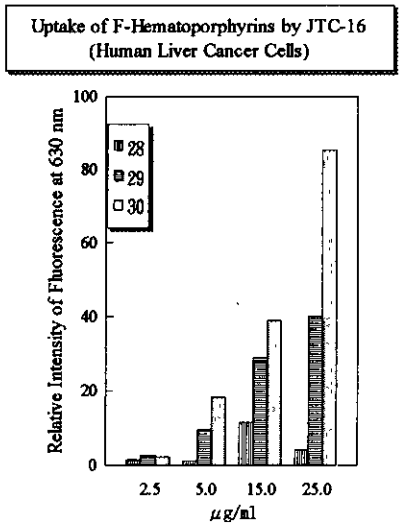
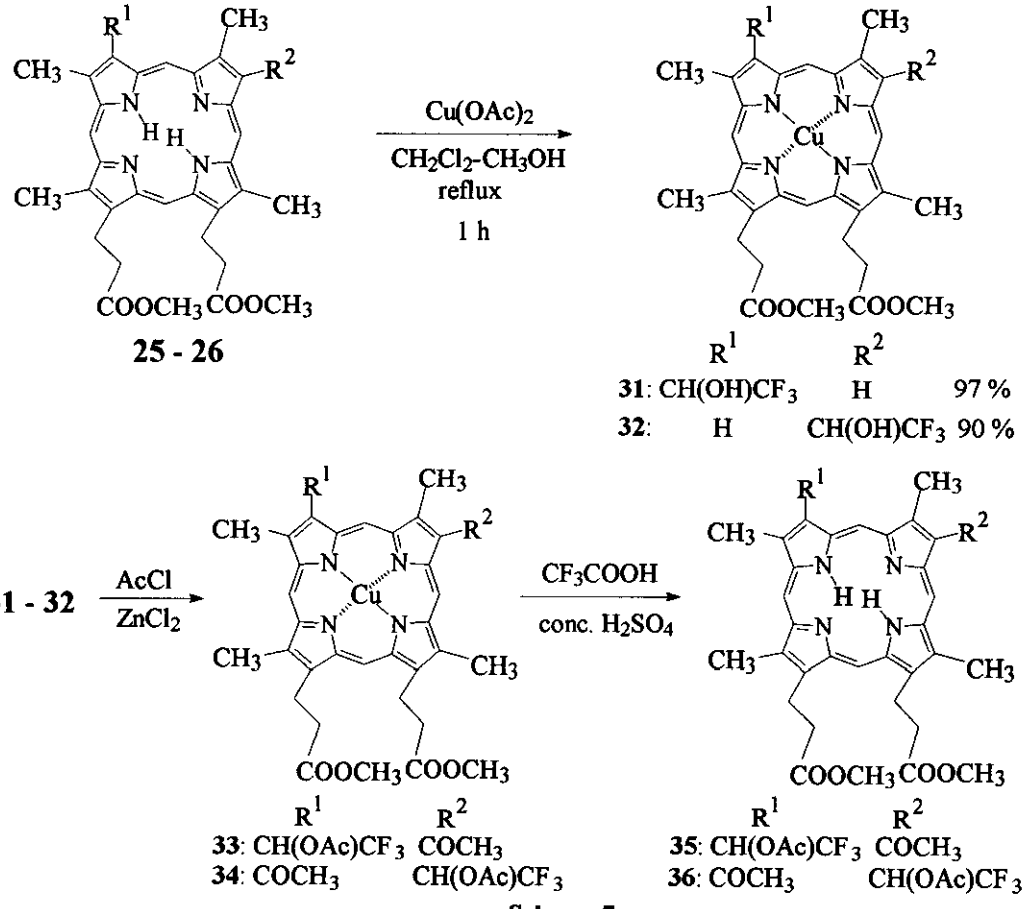


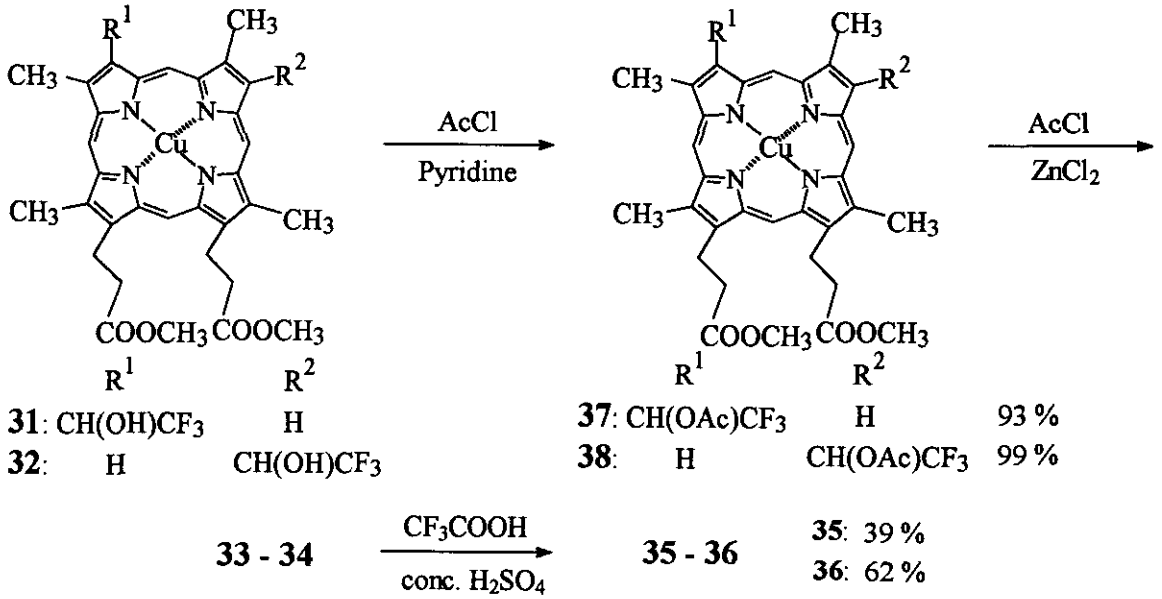
Figure 4



Scheme 7

In these investigation of uptake of TFHE-containing porphyrins to cancer cells, 3,8-TFHE-DPN (**30**), corresponding to hexafluorohematoporphyrin, was found to be taken up more readily than other salts.[21] Thus, we tried to synthesize trifluorohematoporphyrins to clarify the effect of these fluorine substituents. For this purpose, first we investigated acetylation of mono-TFHE compounds (**25** and **26**) and then reduction of the acetyl compounds. In general, the nitrogen atoms of a porphyrin are readily coordinated with metals, and the metalated porphyrin is more stable to acidic conditions. So, mono-TFHE compounds (**25** and **26**) were converted to Cu-complexes to increase the stability of the porphyrin ring, and then subjected to acetylation. Treatment of the mono-TFHE (**25** and **26**) with copper acetate gave Cu-complexes (**31** and **32**) quantitatively. These Cu-complexes (**31** and **32**) reacted with AcCl in CH_2Cl_2 in the presence of $ZnCl_2$, followed by treatment with CF_3COOH-conc. H_2SO_4, to give **35** in 24 % yield. Similarly, **36** was obtained from **32** in 37 % yield, as shown in Scheme 7 on the former page.

During this procedure, two reactions occurred; acetylation of the hydroxyl group of TFHE and acetylation of the porphyrin ring. We thought that the interaction between the OH group of TFHE and $ZnCl_2$ might have decreased the activity of $ZnCl_2$. Therefore, we acetylated the hydroxyl group first, then tried the acetylation of the porphyrin part with $ZnCl_2$. Thus, **31** was treated with pyridine and AcCl to give the acetate (**37**) in 93 % yield, then **37** was treated with $ZnCl_2$ and AcCl, followed by demetalation with CF_3COOH and conc. H_2SO_4 to give **35** in 39 % yield. Thus, the yield was improved only slightly. From Cu-complex (**32**), **36** was obtained in 62 % yield by a similar method. In this case, the yield was improved considerably. In this reaction, the protection of the hydroxyl group improved the acetylation of the ring, effectively (see Scheme 8.)



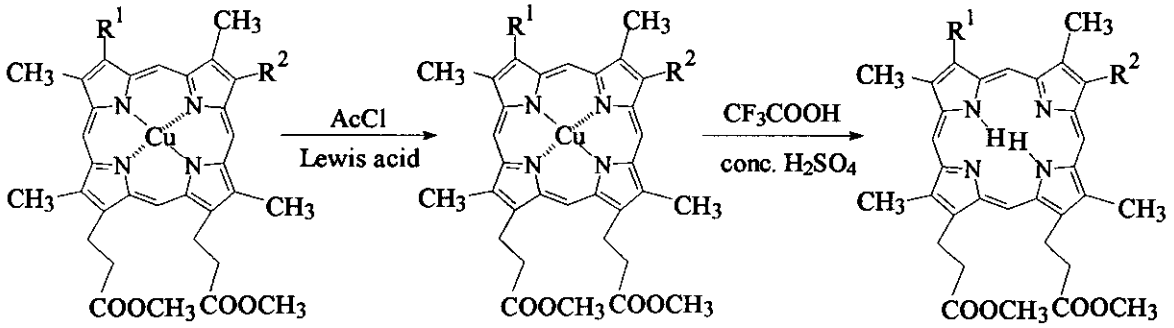
31: R^1 = $CH(OH)CF_3$, R^2 = H
32: R^1 = H, R^2 = $CH(OH)CF_3$
37: R^1 = $CH(OAc)CF_3$, R^2 = H, 93 %
38: R^1 = H, R^2 = $CH(OAc)CF_3$, 99 %
33 - 34
35 - 36
35: 39 %
36: 62 %

Scheme 8

These results suggested that the reactivity of these porphyrins depends on a catalyst and the position of the fluorine containing substituent. So, we examined the reaction with a few catalysts.

When **37** was allowed to react with AcCl in dry CH_2Cl_2 at -50 °C in the presence of $SnCl_4$, followed by treatment with acid to remove copper ion, **35** was obtained in 66 % yield. On the other hand, a similar reaction of **38** gave the demetalated compound of **38** quantitatively. As these results show, the acetylation with $ZnCl_2$ occurred effectively at the 3-position of porphyrin ring, and the acetylation with $SnCl_4$ was more effectively at the 8-position. These results suggest that an electrophilic substitution of a porphyrin ring might occur selectively at a special position depending on a catalyst.

Next, we tried acetylation of **38** in the presence of $TiCl_4$. After demetalation with acids, the product (**36**) was obtained in the 70 % yield. Similarly, **37** was converted to **35**, but the yield was very low (31 %) and a considerable amount of tarry substance was formed. Therefore, this reaction condition is not useful for the acetylation on the 8-position of TFHE-DPDME. In the above reactions, we could obtain 3- and 8-acetylated compounds in 76-80 % by using AcOTFE-porphyrin derivatives (**37** and **38**) (see Scheme 9). During this course, we noticed the difference of reactivity between the 3- and the 8-positions under the reaction condition.



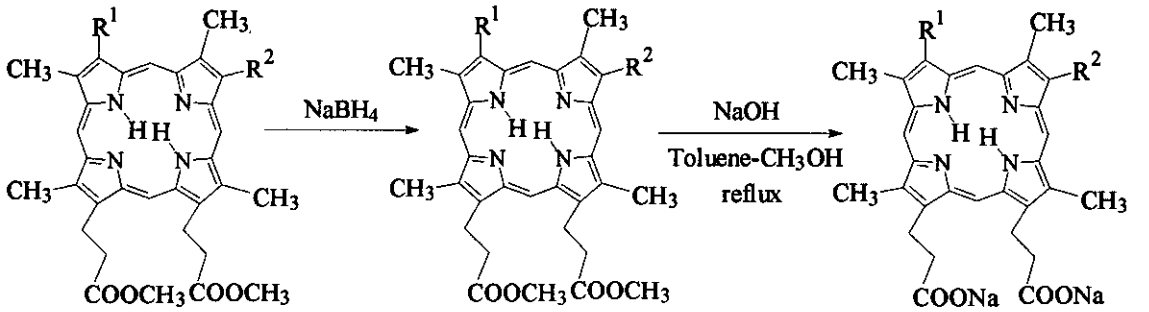
Lewis acid	Condition		Yield (%)	
	Time (h)	Temp.	**35**	**36**
$SnCl_4$	1.0	-50	66	0*
	1.0	25	80	76
$TiCl_4$	1.5	-50	12	0*
	1.5	25	31	70
$ZnCl_2$	2.0	reflux	39	62

Solvent : CH_2Cl_2 * : The demetalated product was obtained quantitatively.

	R^1	R^2
37:	$CH(OAc)CF_3$	H
38:	H	$CH(OAc)CF_3$
33:	$CH(OAc)CF_3$	$COCH_3$
34:	$COCH_3$	$CH(OAc)CF_3$
35:	$CH(OAc)CF_3$	$COCH_3$
36:	$COCH_3$	$CH(OAc)CF_3$

Scheme 9

Acetyl derivative (**35**) was reduced by $NaBH_4$ to give a hydroxyethyl derivative (**39**) in the yield of 85 % as a mixture of diastereomers. Similarly, by the reduction of **36**, **40** was obtained as a mixture of diastereomers (88 %). These were hydrolyzed to sodium salts (**41** and **42**) (see Scheme 10). In a preliminary test of their uptake by human liver cancer cells, hexafluorohematoporphyrin (**30**) was found to be taken up more readily than trifluorohematoporphyrins (**41** and **42**).[22]



35: R^1 = $CH(OAc)CF_3$, R^2 = $COCH_3$
36: R^1 = $COCH_3$, R^2 = $CH(OAc)CF_3$
39: R^1 = $CH(OAc)CF_3$, R^2 = $CH(OH)CH_3$ 85 %
40: R^1 = $CH(OH)CH_3$, R^2 = $CH(OAc)CF_3$ 88 %
41: R^1 = $CH(OH)CF_3$, R^2 = $CH(OH)CH_3$
42: R^1 = $CH(OH)CH_3$, R^2 = $CH(OH)CF_3$

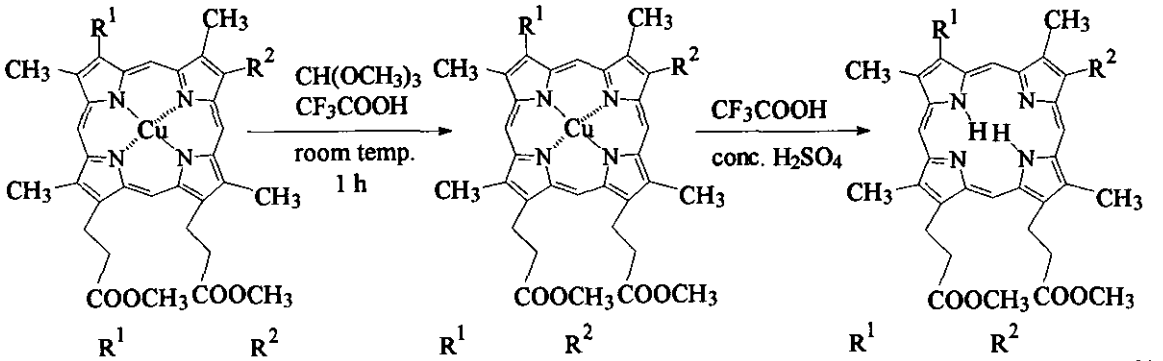
Scheme 10

3. SYNTHESIS OF OTHER TYPES OF FLUORINE CONTAINING PORPHYRIN DERIVATIVES

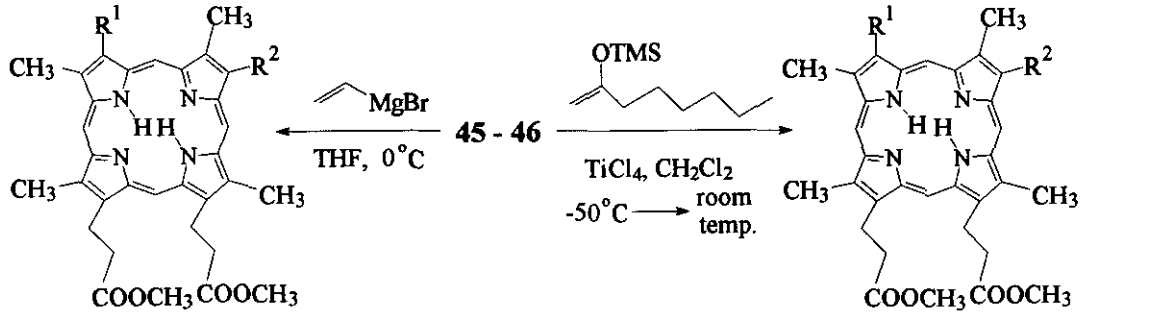
For obtaining fluorine analogs of porphyrin that localize more selectively to a cancer, we tried introduction of other substituents to AcOTFEDPDME-Cu complexes (**37** and **38**).

The formylation of **37** and **38** with trimethyl orthoformate and CF_3COOH gave formyl products (**43** and **44**) in 52 and 51 % yield, respectively. These formyl compounds were treated with CF_3COOH and conc. H_2SO_4 to give demetalated products (**45** and **46**, 93 and 91% yield). Next, we tried the reaction of these formyl compounds (**45** and **46**) with carbanion equivalents such as a Grignard reagent and an enolate ion. Reaction with vinylmagnesium bromide gave allyl alcohols (**47** and **48**) in 85 and 38 % yield, respectively. On the other hand, reaction with 2-trimethylsiloxyoctene in the presence of $TiCl_4$ gave vinyl ketones (**49** and **50**) in 47 and 26 % yields, respectively. These results are shown in Scheme 11.

These compounds (**45** - **50**) were hydrolyzed with sodium hydroxide to sodium salts (**51** - **56**) (see Scheme 12). The localization of these salts to tumor tissue and organs was investigated. The results are shown in Figure 5.[23] These results suggest that some fluorine containing porphyrins would localize to cancer more readily than HpD. Especially, uptakes of those porphyrins which have fluorine substituents at 8-position, such as **52** and **54**, are remarkable. These results suggest that these porphyrins localize to cancer more readily than HpD, and /or localize to liver and kidney less readily than HpD. These porphyrins are taken up specifically and selectively by

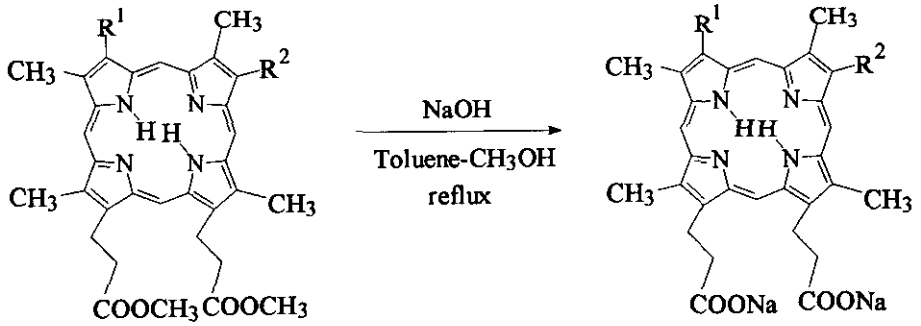


	R^1	R^2					R^1	R^2	
37:	$CH(OAc)CF_3$	H	**43**:	$CH(OAc)CF_3$	CHO	52 %	**45**: $CH(OAc)CF_3$	CHO	93 %
38:	H	$CH(OAc)CF_3$	**44**:	CHO	$CH(OAc)CF_3$	51 %	**46**: CHO	$CH(OAc)CF_3$	91 %



	R^1	R^2					R^1	R^2	
47:	$CH(OAc)CF_3$	$CH(OH)CH=CH_2$	85 %	**49**:	$CH(OAc)CF_3$	$CH=CHCO(CH_2)_5CH_3$	47 %		
48:	$CH(OH)CH=CH_2$	$CH(OAc)CF_3$	38 %	**50**:	$CH=CHCO(CH_2)_5CH_3$	$CH(OAc)CF_3$	26 %		

Scheme 11



	R^1	R^2		R^1	R^2
45:	$CH(OAc)CF_3$	CHO	**51**:	$CH(OH)CF_3$	CHO
46:	CHO	$CH(OAc)CF_3$	**52**:	CHO	$CH(OH)CF_3$
47:	$CH(OAc)CF_3$	$CH(OH)CH=CH_2$	**53**:	$CH(OH)CF_3$	$CH(OH)CH=CH_2$
48:	$CH(OH)CH=CH_2$	$CH(OAc)CF_3$	**54**:	$CH(OH)CH=CH_2$	$CH(OH)CF_3$
49:	$CH(OAc)CF_3$	$CH=CHCO(CH_2)_5CH_3$	**55**:	$CH(OH)CF_3$	$CH=CHCO(CH_2)_5CH_3$
50:	$CH=CHCO(CH_2)_5CH_3$	$CH(OAc)CF_3$	**56**:	$CH=CHCO(CH_2)_5CH_3$	$CH(OH)CF_3$

Scheme 12

cancer cells. We are now investigating the biological behavior of these porphyrins more extensively and synthesizing other porphyrin derivatives.

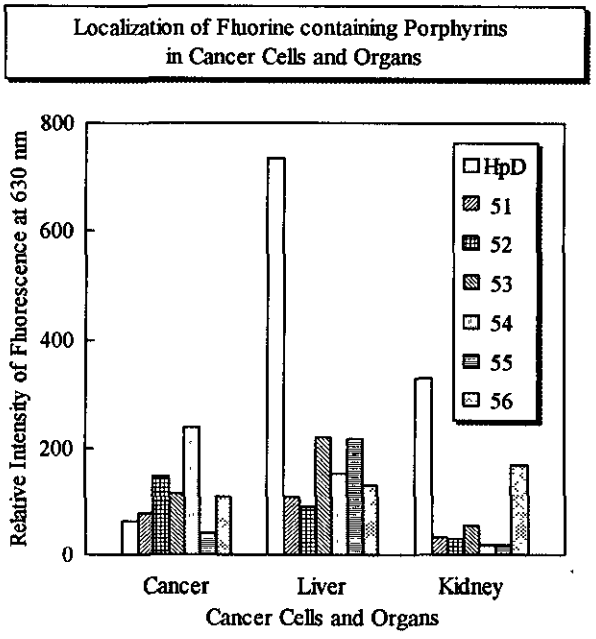


Figure 5

4. CONCLUDING REMARKS

We have synthesized fluorine analogs of naturally important porphyrins. Some of them are taken up by special kinds of tumor cells selectively. Further, modification of the vinyl substituents at 3- or 8-position by a substituent containing fluorine atoms and a nonfluorinated substituent showed similar effects. A fluorine-containing substiutuent on the 8-position showed a larger effect. These results are obtained *in vitro*. Now, we are planning *in vivo* experiments. If some of these localize to a special cancer selectively, they will be very useful for diagnosis and therapy of cancers and replace HpD's, since they are complex mixtures and the component which localizes is not determined. Our compounds are chemically pure and will give more reliable results than HpD's.

REFERENCES AND NOTES

1. 'Porphyrins and metalloporphyrins', K. M. Smith (ed) Elsevier Scientific Publishing Company, Amsterdam, 1975.
2. Concerning chemistry and biochemistry of HpD, see 'Advances in Experimental Medicine and Biology', **160**, 'Porphyrin Photosensitization', D. Kessel and T. J. Dougherty (eds.) Plenum, New York, 1983.
3. W. Hausmann, *Biochem. Z.*, 1911, **30**, 276.
4. D. Kessel, *Biochem. Pharmacology*, 1984, **33,** 1389.
5. R. K. Pandey, F. -Y. Shiau, T. J. Dougherty, and K. M. Smith, *Tetrahedron*, 1991, **47**, 9571.
6. K. Aizawa, T. Okunaka, T. Ohtani, H. Kawabe, Y. Yasunaka, S. O'hata, H. Kato, Y. Hayata, and T. Saito, *Photochem. Photobiol.*, 1987, **46**, 789.
7. N. Miyoshi, A. Hirata, K. Kunimi, K. Sakurai, K. Sakamoto, N. Matumoto, H. Hisazumi, and M. Fukuda, *Lasers Med. Sci.*, 1988, **3**, 185.

8. S. Nakajima, H. Hayashi, K. Ohshima, K. Yamazaki, Y. Kubo, N. Samejima, Y. Kakiuchi, Y. Shindoh, H. Koshimizu, I. Sakata, and H. Yamauchi, *Photochem. Photobiol.*, 1987, **46**, 783.

9. a. R. K. Pandey, D. A. Bellnier, K. M. Smith, and T. J. Dougherty, *Photochem. Photobiol.*, 1991, **53**, 65. b. S. Chernomorsky, C. Wong, and R. D. Poretz, *Photochem. Photobiol.*, 1992, **55**, 205.

10. N. Miyoshi, H. Hisazumi, and M. Fukuda, *Photomed. Photobiol.*, 1988, **10**, 69.

11. M. Kuwabara, T. Yamamoto, O. Inanami, and F. Sato, P*hotochem. Photobiol.*, 1989, **49**, 37.

12. H. Sato, M. Nishi, and I. Kumadaki, *Chemistry Express*, 1988, **3**, 183.

13. H. Sato, R. Tsujimoto, I. Kumadaki, M. Nishi, K. Ido, and K. Kimura, *J. Jpn. Soc., Laser Medicine*, 1986, **6**, 95

14. Concerning the biological effects of fluorine compounds, see 'Biomedicinal Aspects of Fluorine Chemistry', R. Filler and Y. Kobayashi (Eds), Kodansha, Tokyo, 1983.

15. H. H. Inhoffen, H. Brockmann Jr., and K.-M. Bliesener, *Liebigs Ann. Chem.*, 1969, **730**,173.

16. a. A. Ando, T. Shinada, S. Kinoshita, N. Arimura, M. Koyama, T. Nagai, T. Miki, I. Kumadaki, and H. Sato, *Chem. Pharm. Bull.*, 1990, **38**, 2175. b. A. Ando, T. Shinada, S. Kinoshita, N. Arimura, T. Nagai, T. Miki, and I. Kumadaki, *J. Fluorine Chemistry*, 1989, **42**, 293.

17. F. Sparatore and D. Mauzerall, *J. Org. Chem.*, 1960, **25**, 1073.

18. a. W. S. Caughey, J. O. Alben, W. Y. Fujimoto, and J. L. York, *J. Org. Chem.*, 1966, **31**, 2631. b. R. K. Dinello and D. H. Dolphin, *J. Org. Chem.*, 1981, **46**, 3498.

19. A. Guy, A. Lobgeois, and M. Lemaire, *J. Fluorine Chem.*, 1986, **32**, 361.

20. Efficiency of this solvent system for extraction of porphyrins is shown by H. Sato, K. Ido, and K. Kimura, *Clin. Chem.*, 1994, **40**, 1239.

21. a. A. Ando, T. Kitamura, S. Aono, H. Sato, M. Omote, M. Koyama, T. Takagi, T. Miki, I. Kumadaki, and H. Sato, *Heterocycles*, 1993, **35**, 1309. b. A. Ando, M. Omote, and I. Kumadaki, *Porphyrins*, 1992, **1**, 243.

22. M. Omote, A. Ando, M. Koyama, T. Takagi, and I Kumadaki, *Heterocycles* ,1994, **39**, 381.

23. M. Omote, A. Ando, T. Takagi, M. Koyama, I. Kumadaki, and H. Sato, *Chem. Pharm. Bull.*, submitted.

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