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Novel Chemoselective De-esterification of Esters of Polyacetoxy Aromatic Acids by Lipases#

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Abstract: *Candida cylindracea* lipase (CCL) and porcine pancreatic lipase (PPL) have been used for deacetylation of peracetates of methyl and ethyl esters of six different polyphenolic acids in organic solvents. Exclusive de-esterification of the ester groups derived from the phenolic hydroxy and aliphatic acid over the ester group of the aromatic acid and aliphatic alcohol has been achieved affording the corresponding esters of phenolic acids in as high yields as 90-97%. The results have been corroborated with the mechanism of lipase action.

INTRODUCTION

Search for reactions that show greater selectivity and are environmentally-friendly has resulted in the development of enzyme- and microorganism-catalysed synthesis. Thus, lipases and proteases have found increasing use for selective protection and deprotection of hydroxy, amino and carboxy groups in organic synthesis since these enzymes work under mild and near-neutral conditions in the presence of both base- and acid- sensitive functional groups and they are highly selective.[1] We have earlier demonstrated the capabilities of lipases from porcine pancreas (PPL) and *Candida cylindracea* (CCL) in regioselective deacetylation of the acetoxy group(s) in different classes of peracetylated polyphenolic compounds and it was observed that all acetoxy group(s) except the ones at the *ortho* or *peri* position(s) to the nuclear carbonyl group are deacetylated.[2-7] We postulated that the nuclear carbonyl group present in the substrate is engaged in the formation of a transient (dynamic) Schiff's base type complex with the free ξ-amino function of the lysine residue present in the active site of PPL (an analogy to the human pancreatic lipase). The formation of this complex causes the *ortho* or *peri* acetoxy function to be embedded under the hydrophobic bulk of the active site of the enzyme and the serine -OH takes part in deacetylation of other more suitably placed acetoxy function(s) in the same molecule.[6] However, no concrete proof for this mechanism could be given as the active site structure of PPL is not known and also because the Schiff's base formation, being a transient (dynamic) process, could not be verified.

Aromatic hydroxy acids and their esters form an important class of natural products and play a significant role in the biosynthesis of other important polyphenolic compounds.[8] Esters do not form Schiff's bases easily, and therefore may be used as chemical probes for studying the mechanism of lipase action in the enzymatic deacetylation

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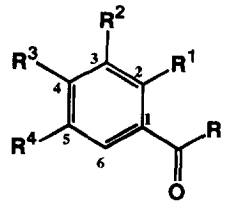
studies discussed above. It is expected that deacetylation reactions on the esters of polyacetoxy aromatic acids catalysed by lipases would not involve formation of Schiff's base complexes and as a result random orientation of different acetoxy groups in the active site of the lipase might result in deacetylation of the *ortho* acetoxy group also, resulting in the formation of an *ortho* hydroxy product together with other possible product(s). However, as per our scrutiny of literature, esters of polyacetoxy aromatic acids have not been subjected to enzymatic hydrolysis/ deacetylation studies. With an aim to substantiate our earlier proposed mechanism[6] of lipase action during deacetylation of peracetylated polyphenolics and to study the biotransformations on the esters of polyacetoxy aromatic acids, we wish to report here deacetylation studies on methyl and ethyl esters of six different polyacetoxybenzoic acids, as well as on two diacetoxy aromatic ketones as model compounds.



1 R=OCH_3, R^1=R^2=R^4=H, R^3=$OCOCH_3$

2 R=OCH_2CH_3, R^1=R^2=R^4=H, R^3=$OCOCH_3$

3 R=OCH_3, R^1=$OCOCH_3$, R^2=R^3=R^4=H

4 R=OCH_2CH_3, R^1=$OCOCH_3$, R^2=R^3=R^4=H

5 R=OCH_3, R^1=R^3=$OCOCH_3$, R^2=R^4=H

6 R=OCH_2CH_3, R^1=R^3=$OCOCH_3$, R^2=R^4=H

7 R=OCH_3, R^1=R^3=H, R^2=R^4=$OCOCH_3$

8 R=OCH_2CH_3, R^1=R^3=H, R^2=R^4=$OCOCH_3$

9 R=OCH_3, R^1=R^2=R^3=$OCOCH_3$, R^4=H

10 R=OCH_2CH_3, R^1=R^2=R^3=$OCOCH_3$, R^4=H

11 R=OCH_3, R^1=H, R^2=R^3=R^4=$OCOCH_3$

12 R=OCH_2CH_3, R^1=H, R^2=R^3=R^4=$OCOCH_3$

13 R=CH_3, R^1=R^3=$OCOCH_3$, R^2=R^4=H

14 R=C_6H_5, R^1=R^3=$OCOCH_3$, R^2=R^4=H

15 R=OCH_3, R^1=R^2=R^4=H, R^3=OH

16 R=OCH_2CH_3, R^1=R^2=R^4=H, R^3=OH

17 R=OCH_3, R^1=OH, R^2=R^3=R^4=H

18 R=OCH_2CH_3, R^1=OH, R^2=R^3=R^4=H

19 R=OCH_3, R^1=$OCOCH_3$, R^2=R^4=H, R^3=OH

20 R=OCH_3, R^1=R^3=OH, R^2=R^4=H

21 R=OCH_2CH_3, R^1=$OCOCH_3$, R^2=R^4=H, R^3=OH

22 R=OCH_2CH_3, R^1=R^3=OH, R^2=R^4=H

23 R=OCH_3, R^1=R^3=H, R^2=$OCOCH_3$, R^4=OH

24 R=OCH_3, R^1=R^3=H, R^2=R^4=OH

25 R=OCH_2CH_3, R^1=R^3=H, R^2=$OCOCH_3$, R^4=OH

26 R=OCH_2CH_3, R^1=R^3=H, R^2=R^4=OH

27 R=OCH_3, R^1=OH, R^2=R^3=$OCOCH_3$, R^4=H

28 R=OCH_3, R^1=R^2=R^3=OH, R^4=H

29 R=OCH_2CH_3, R^1=OH, R^2=R^3=$OCOCH_3$, R^4=H

30 R=OCH_2CH_3, R^1=R^2=R^3=OH, R^4=H

31 R=OCH_3,R^1=H, R^2=R^4=$OCOCH_3$, R^3=OH

32 R=OCH_3, R^1=H, R^2=R^3=R^4=OH

33 R=OCH_2CH_3, R^1=H, R^2=R^4=$OCOCH_3$, R^3=OH

34 R=OCH_2CH_3, R^1=H, R^2=R^3=R^4=OH

35 R=CH_3, R^1=$OCOCH_3$, R^2=R^4=H, R^3=OH

36 R=CH_3, R^1=R^3=OH, R^2=R^4=H

37 R=C_6H_5, R^1=$OCOCH_3$, R^2=R^4=H, R^3=OH

38 R=C_6H_5, R^1=R^3=OH, R^2=R^4=H

RESULTS AND DISCUSSION

The deacetylation reactions on esters **1-12** and ketones **13-14** have been carried out with PPL and CCL in dry organic solvents (tetrahydrofuran and diisopropyl ether) containing added *n*-butanol. The results of CCL- and PPL- catalysed deacetylation reactions are summarised in Tables 1 and 2, respectively; without addition of the lipase these reactions did not indicate any deacetylation. A comparison of CCL and PPL induced deacetylation

reactions on peracetylated phenolic acids esters **1-12** revealed that the reactions catalysed by CCL in diisopropyl ether were faster in comparison to those catalysed by PPL in THF. However, no clear cut regioselectivity was observed with any of the enzymes.

Table 1. Deacetylation reactions catalysed by *Candida cylindracea* lipase (CCL) on esters of polyacetoxy aromatic acids and diacetoxy aromatic ketones in dry diisopropyl ether at 28-30°C.

Substrate	**Reaction time(hrs)**	**Product(s) (% yield)**
Methyl 4-acetoxybenzoate (**1**)[12]	16	Methyl 4-hydroxybenzoate (**15**)[9c] (95)
Ethyl 4-acetoxybenzoate (**2**)[12]	14	Ethyl 4-hydroxybenzoate (**16**)[9c] (97)
Methyl 2-acetoxybenzoate (**3**)[9a]	72	Methyl 2-hydroxybenzoate (**17**)[9d] (90)
Ethyl 2-acetoxybenzoate (**4**)[9a]	48	Ethyl 2-hydroxybenzoate (**18**)[9d] (94)
Methyl 2,4-diacetoxybenzoate (**5**)[10]	18	Methyl 2-acetoxy-4-hydroxybenzoate (**19**) (48) *and* Methyl 2,4-dihydroxybenzoate (**20**)[9c] (47)
Ethyl 2,4-diacetoxybenzoate (**6**)	18	Ethyl 2-acetoxy-4-hydroxybenzoate (**21**) (50) *and* Ethyl 2,4-dihydroxybenzoate (**22**)[11] (46)
Methyl 3,5-diacetoxybenzoate (**7**)	5	Methyl 3-acetoxy-5-hydroxybenzoate (**23**) (30) *and* Methyl 3,5-dihydroxybenzoate (**24**)[11] (65)
Ethyl 3,5-diacetoxybenzoate (**8**)	3	Ethyl 3-acetoxy-5-hydroxybenzoate (**25**) (32) *and* Ethyl 3,5-dihydroxybenzoate (**26**)[11] (65)
Methyl 2,3,4-triacetoxybenzoate (**9**)	120	Methyl 3,4-diacetoxy-2-hydroxybenzoate (**27**) (60) *and* Methyl 2,3,4-trihydroxybenzoate (**28**)[9f] (20)
Ethyl 2,3,4-triacetoxybenzoate (**10**)	48	Ethyl 3,4-diacetoxy-2-hydroxybenzoate (**29**) (62) *and* Ethyl 2,3,4-trihydroxybenzoate (**30**)[9f] (24)
Methyl 3,4,5-triacetoxybenzoate (**11**)[13]	10	Methyl 3,5-diacetoxy-4-hydroxybenzoate (**31**) *and* Methyl 3,4,5-trihydroxybenzoate (**32**)[9b] (12)
Ethyl 3,4,5-triacetoxybenzoate (**12**)[9b]	10	Ethyl 3,5-diacetoxy-4-hydroxybenzoate (**33**) *and* Ethyl 3,4,5-trihydroxybenzoate (**34**)[9b] (11)
2,4-Diacetoxyacetophenone (**13**)[9g]	11	2-Acetoxy-4-hydroxyacetophenone (**35**)[2] (50) *and* 2,4-Dihydroxyacetophenone (**36**)[9g] (40)
2,4-Diacetoxybenzophenone (**14**)[9h]	40	2-Acetoxy-4-hydroxybenzophenone (**37**)[7] (35) *and* 2,4-Dihydroxybenzophenone (**38**)[9h] (50)

The methyl and ethyl monoacetoxybenzoates **1-4** on treatment with CCL yielded the corresponding hydroxy esters **15-18**, respectively in very high yields (90-97%) in a chemoselective mode, irrespective of the position of the acetoxy group with respect to the nuclear alkoxycarbonyl group (Table 1). In a similar fashion, the

diacetoxybenzoates **5-8** underwent deacetylation at all three positions, *i.e. ortho, meta* and *para* to give mono- and dihydroxybenzoates **19-26.** The kinetics of the reaction suggested that in methyl or ethyl 2,4-diacetoxybenzoates **5** and **6,** the *para* acetoxy function undergoes deacetylation faster as compared to the *ortho* acetoxy group to give initially the compounds **19** and **21,** respectively; however complete deacetylation occurred when the reaction time was sufficiently prolonged to give the corresponding dihydroxy products **20** and **22.** The structures of compounds **19** and **21** were assigned on the basis that none of the compounds showed a colour reaction with alcoholic ferric chloride solution. Further no peak was observed for the chelated hydroxyl group in their respective ^{1}H NMR spectrum. Interestingly, in the symmetric ethyl and methyl 3,5-diacetoxybenzoates **7** and **8,** in addition to the dihydroxybenzoates **24** and **26,** the monoacetoxy products **23** and **25** were obtained when the reaction was quenched in the initial stages. It may be noted that the partially acetylated compounds like **23** and **25** are difficult to prepare by purely chemical methods, as illustrated by the recently published cumbersome synthesis of **23.**[15]

Table 2. Deacetylation reactions catalysed by porcine pancreatic lipase (PPL) on esters of polyacetoxy aromatic acids and diacetoxy aromatic ketones in THF at 28-30°C.

Substrate	**Reaction time**	**Product(s) (% yield)**
Methyl 4-acetoxybenzoate (**1**)[12]	5 days	Methyl 4-hydroxybenzoate (**15**)[9c] (80)
Ethyl 4-acetoxybenzoate (**2**)[12]	5 days	Ethyl 4-hydroxybenzoate (**16**)[9c] (85)
Methyl 2,4-diacetoxybenzoate (**5**)[10]	23 days	Methyl 2-acetoxy-4-hydroxybenzoate (**19**) (90) *and* Methyl 2,4-dihydroxybenzoate (**20**)[9c] (5)
Ethyl 2,4-diacetoxybenzoate (**6**)	22 days	Ethyl 2-acetoxy-4-hydroxybenzoate (**21**) (87) *and* Ethyl 2,4-dihydroxybenzoate (**22**)[11] (6)
Methyl 3,5-diacetoxybenzoate (**7**)	2 days	Methyl 3-acetoxy-5-hydroxybenzoate (**23**) (31) *and* Methyl 3,5-dihydroxybenzoate(**24**)[11] (60)
Ethyl 3,5-diacetoxybenzoate (**8**)	2 days	Ethyl 3-acetoxy-5-hydroxybenzoate (**25**) (32) *and* Ethyl 3,5-dihydroxybenzoate (**26**)[11] (65)
Methyl 2,3,4-triacetoxybenzoate (**9**)	30 days	Methyl 3,4-diacetoxy-2-hydroxybenzoate (**27**) (50) *and* Methyl 2,3,4-trihydroxybenzoate (**28**)[9f] (22)
Ethyl 2,3,4-triacetoxybenzoate (**10**)	19 days	Ethyl 3,4-diacetoxy-2-hydroxybenzoate (**29**) (60) *and* Ethyl 2,3,4-trihydroxybenzoate (**30**)[9f] (25)
Ethyl 3,4,5-triacetoxybenzoate (**12**)[9b]	5 days	Ethyl 3,5-diacetoxy-4-hydroxybenzoate (**33**) *and* Ethyl 3,4,5-trihydroxybenzoate (**34**)[9b] (34)
2,4-Diacetoxyacetophenone (**13**)[9g]	30 hrs	2-Acetoxy-4-hydroxyacetophenone (**35**)[2] (5)
2,4-Diacetoxyacetophenone* (**13**)[9g]	40 hrs	2-Acetoxy-4-hydroxyacetophenone (**35**)[2] (40)
2,4-Diacetoxybenzophenone (**14**)[9h]	40 hrs	2-Acetoxy-4-hydroxybenzophenone (**37**)[7] (30)
2,4-Diacetoxybenzophenone* (**14**)[9h]	40 hrs	2-Acetoxy-4-hydroxybenzophenone (**37**)[7] (80)

* in diisopropyl ether

The methyl and ethyl 2,3,4-triacetoxybenzoates **9** and **10** under these reaction conditions resulted in the formation of a number of products as revealed by TLC examination. Methyl 2,3,4-triacetoxybenzoate (**9**) yielded a mixture; two compounds, *i.e.* methyl 3,4-diacetoxy-2-hydroxybenzoate (**27**) and methyl 2,3,4-trihydroxybenzoate (**28**) could be isolated by using preparative TLC, the latter compound was identified by its comparison with an authentic sample. Methyl 3,4-diacetoxy-2-hydroxybenzoate (**27**) was identified from its spectral data; in its EIMS, the molecular ion peak appeared at *m/z* 268 indicating the presence of two acetoxy groups, which was further supported by its NMR spectra as the two acetoxy groups appeared as two singlets (each for three protons) at δ 2.26 and 2.28 in its ^{1}H NMR and at δ 19.89 and 20.17 in its ^{13}C NMR spectrum. The presence of a peak at δ 10.76 in its ^{1}H NMR spectrum indicated the presence of a chelated hydroxyl group, which was confirmed by the observance of a bathochromic shift of 29 nm in the λmax of **27** in the presence of $AlCl_3$ shift reagent, it is reported[16] that $AlCl_3$ induces a bathochromic shift of 16-30 nm in the UV absorption maxima of compounds having a chelated hydroxyl group. In addition, **27** gave a black colour with alcoholic ferric chloride solution which further substantiated the presence of a chelated hydroxyl group. Thus compound **27** was assigned as methyl 3,4-diacetoxy-2-hydroxybenzoate.

In a similar fashion, ethyl 2,3,4-triacetoxybenzoate (**10**) yielded a mixture from which only two compounds, *i.e.* ethyl 3,4-diacetoxy-2-hydroxybenzoate (**29**) and ethyl 2,3,4-trihydroxybenzoate (**30**) could be isolated by preparative TLC. Compound **30** was identified by comparison with its authentic sample and the constitution of compound **29** was assigned from its spectral data and colour reactions. The observance of a molecular ion peak at *m/z* 282 in its EIMS indicated the presence of two acetoxy groups, which was confirmed by singlets at δ 2.36 and 2.38 in its ^{1}H NMR and at δ 20.27 and 20.70 in its ^{13}C NMR spectrum. A bathochromic shift of 20 nm in its λmax in the presence of $AlCl_3$ shift reagent indicated the presence of a chelated hydroxyl group,[16] which was further substantiated by its colour reaction with alcoholic ferric chloride. Thus compound **29** was assigned as ethyl 3,4-diacetoxy-2-hydroxybenzoate. It may be mentioned that the partially protected trihydroxybenzoates **27** and **29** are difficult to obtain in as high yields as 60% by purely chemical reactions.

Methyl 3,4,5-triacetoxybenzoate **11** on enzymatic deacetylation yielded a mixture, from which only the completely deacetylated compound **32** could be isolated in the pure form by preparative TLC. The HPLC examination of the remaining product mixture on a reverse-phase column (Shim-pack CLC-ODS(M), 25 cm) using water-methanol (3:7) as the solvent at the flow rate of 0.75 ml/min and monitored at λ 254 nm revealed it to be a mixture of two compounds, *i.e.* the starting triacetoxybenzoate **11** and a partially deacetylated compound having retention times 5.60 and 4.49 min, respectively. However repeated attempts to isolate the two components in the pure form by preparative HPLC, column chromatography or preparative TLC failed. Surprisingly, the mixture was found to melt sharply at 108-09°C and in its ^{1}H NMR spectrum, the aromatic protons H-2 and H-6 for both compounds appeared at the same position[18]. Similarly in its ^{13}C NMR spectrum, both C-2 and C-6 for the two compounds appeared at δ 121.90 and C-3 and C-5 also appeared at the same position[18]. Thus it seems that there are two symmetrical compounds in the mixture, *i.e.* **11** and methyl 3,5-diacetoxy-4-hydroxybenzoate (**31**). However, there appeared two singlets in the ^{1}H NMR spectrum of the mixture at δ 2.29 and 2.33 having the intensity in the ratio 2:1 for the acetoxyl protons,[18] while in the starting triacetoxy compound **11** the three acetoxyl groups appeared as a singlet at δ 2.25. The relative integrals for the acetoxyl protons of the two compounds suggested that **11** and **31** are present in the ratio 4:3 in the mixture.

Ethyl 3,4,5-triacetoxybenzoate **12** under similar reaction conditions also yielded a mixture, from which only the completely deacetylated compound **34** could be isolated in the pure form by preparative TLC. The other

product was again found to be an inseparable mixture. However, HPLC examination (reverse phase column, Shim-pack CLC-ODS(M), 25cm) using water-methanol (3:7) as the solvent at the flow rate of 0.75 ml/min and monitored at λ 254 nm revealed it to be a mixture of two compounds, *i.e.* the starting triacetoxybenzoate **12** and a partially deacetylated compound having retention times 6.83 and 5.07 min, respectively. The mixture was found to melt sharply at 140 °C and in its ^{1}H NMR spectrum, the aromatic protons H-2 and H-6 for both compounds appeared at same position[18]. Similarly in its ^{13}C NMR spectrum, both C-2 and C-6 appeared at δ 121.77 and C-3 and C-5 for both compounds likewise appeared at the same position[18]. Thus both compounds of the mixture are symmetrical. Furthermore, the ^{1}H NMR spectrum of the mixture exhibited two singlets for the acetoxyl protons at δ 2.30 and 2.33 integrating in the ratio 2:1, whereas the starting triacetoxy compound **12** exhibited a singlet at δ 2.28 for the three acetoxyl groups. The other compound was, therefore inferred to be the one formed by monodeacetylation at the C-4 position and was thus assigned the structure ethyl 3,5-diacetoxy-4-hydroxybenzoate (**33**). The relative integration for the acetoxyl protons suggested that the compounds **12** and **33** are present in the ratio 4:3 in the biotransformation mixture.

Though we have earlier reported highly regioselective deacetylation reactions on peracetylated polyphenolic compounds with two lipases (PPL and CCL), we have not categorically differentiated the regioselectivity exhibited by PPL and CCL with different aryl-alkyl ketones[2-7]. Therefore, to further confirm our previous results[2-7] on regioselective deacetylations in polyacetoxy aryl-alkyl ketones, we have now repeated deacetylation experiments with two representative peracetylated aromatic ketones *viz.* **13** and **14** using PPL and CCL in different solvent systems. The deacetylation results show that PPL displays a high order of regioselectivity in both THF and diisopropyl ether, as deacetylation occurs selectively at the *para* position (Table 2). No such selectivity was observed with CCL as deacetylation occurs at *para* as well as at the *ortho* position (Table 1). The lipase PPL is a mammalian enzyme and could be similar to human pancreatic lipase (whose structure is known and it possesses a lysine residue in the active site), PPL is asumed to likewise possess lysine residue in its active site which forms a Schiff's base with the carbonyl group of ketones, the formation of which in turn is responsible for the regioselective deacetylation of polyacetoxy aromatic ketones as suggested earlier by us.[6] On the other hand, CCL is a microbial lipase, which perhaps lacks the lysine residue in its active site and is unable to form Schiff's bases with the carbonyl group of the aryl-alkyl ketones. Hence, a similar selectivity is not observed with CCL. The lack of recognition of different acetoxy functions in esters **1-12** by both PPL and CCL, when compared with the results obtained in ketones probably stems from the inability of the esters to form Schiff's bases with the ξ-amino function of the lysine residue in the active site of the porcine pancreatic lipase. As a result, the random placement of different acetoxy functions in the active site of the lipase results in deacetylation of all acetoxy functions. Thus, methyl and ethyl 2,3,4-triacetoxybenzoates **9** and **10** underwent deacetylation of the *ortho* acetoxy function with PPL to yield the compounds **27** and **28**, respectively (Table 2) in as high as 50-60% yields. Similarly, the 2,4-diacetoxybenzoic acid esters **5** and **6** underwent deacetylation at both the *ortho* and *para* acetoxy functions with PPL to yield the corresponding dihydroxybenzoic acid esters **20** and **22**, respectively (Table 2). As reported by us earlier, no deacetylation of *ortho* acetoxy function occurs on incubating either 2,4-diacetoxy- and 2,3,4-triacetoxyacetophenone with PPL,[2] or 2,3,4-triacetoxypropiophenone with PPL.[7] These findings substantiate our hypothesis of the presence of lysine in the active site of the procine pancreatic lipase and the formation of the Schiff's base type complexes during deacetylation of peracetates of polyphenolic compounds bearing nuclear (keto) carbonyl group.

To conclude, the present study has shown interesting and potentially useful chemoselectivity during lipase-

catalysed deacetylation reactions in dry organic solvents on peracetates of esters of polyphenolic acids. Both *Candida cyclindracea* lipase (CCL) and porcine pancreatic lipase (PPL) in dry organic solvents perform exclusively and efficiently de-esterification of the ester groups formed from phenolic hydroxyl group and aliphatic acid over the one obtained from the aromatic acid and aliphatic alcohol in all the twelve substrates that have been studied. As it is difficult to selectively hydrolyse these two types of ester groups by chemical methods, the biocatalytic reactions reported here should find utility in the selective protection of hydroxyaromatic carboxylic acids. This study also supports the hypothesis of Schiff's base formation during deacetylation reactions catalysed by PPL in polyphenolic aromatic ketones and their analogs.[6] Additionally, eleven new compounds, *i.e.* **6, 7, 8, 9, 10, 19, 21, 23, 25, 27** and **29** have been prepared. The partially acetylated compounds **23, 25, 27** and **29** could find use in the synthesis of new analogs of different classes of bioactive naturally occurring polyphenolic compounds. Selected spectral data, not reported earlier for twenty three known compounds together with spectral data for the eleven new compounds have also been included. Physical and spectral data of **35** and **37** have been published by us earlier[2,7] and data for the inseparable mixtures obtained in deacetylation reactions on 3,4,5-triacetoxybenzoates **11** and **12** is given in the 'References and Notes' section.[18]

EXPERIMENTAL

Melting points were taken in a sulphuric acid bath and are uncorrected. The ^{1}H and ^{13}C NMR spectra were recorded either on Perkin-Elmer R-32 (90 MHz) spectrometer or on a Jeol JNM FX-200 FT NMR (250 MHz) spectrometer and UV spectra on a Perkin-Elmer model 554 spectrophotometer. The IR spectra were recorded on a Shimadzu model 435 spectrophotometer, the HPLC analysis was performed on a Shimadzu model LC-10A chromatograph, and the mass spectra were recorded on a Varian-Mat 311A or on a Jeol AX505W instrument.

Materials: The lipases from *Candida cylindracea* (CCL, Type VII) and porcine pancreas (PPL, Type II) were purchased from Sigma Chemical Company (USA) and used after keeping *in vacuo* over P_2O_5 for 24 hrs. Tetrahydrofuran and diisopropyl ether were distilled, dried and stored over molecular sieves (4A°), while *n*-butanol was distilled and dried over ignited potassium carbonate.

Preparation of substrates. The hydroxy benzoates were prepared by refluxing the corresponding hydroxy acid in excess methyl or ethyl alcohol in the presence of a few drops of concentrated H_2SO_4. On completion of the reaction (30-40 hrs), excess alcohol was removed and the residue poured over crushed ice affording the hydroxy ester as a white solid. Peracetates of the hydroxy benzoates and aromatic ketones were prepared by acetic anhydride/H_2SO_4 method either at room temperature or at 80°C. All peracetates were fully characterised prior to their use.

Lipase-catalysed deacetylation. The peracetylated substrate (2-3 mmol) was dissolved in the organic solvent (THF or diisopropylether, 20-30 ml) containing *n*-butanol (4-6 mol eq.) and lipase powder (200 mg) was added. The suspension was stirred at 28-30°C and progress of the reaction was monitored by TLC on precoated Merck silica gel plates. The reaction was quenched by filtering off the lipase. The solvent was evaporated to dryness *in vacuo* and the product(s) purified by preparative TLC and/or column chromatography, and fully characterised from their physical and spectral data and by a comparison with the reported data, if possible. [9-14]

Methyl 4-acetoxybenzoate (**1**). White solid, mp 78°C (lit[12] mp 78-80°C); UV(MeOH): 242 nm; ^{1}H NMR($CDCl_3$): δ 2.30(3H, s, $OCOCH_3$), 3.81(3H, s, $COOCH_3$), 6.90(2H, d, *J*= 8Hz, H-3 and H-5), 7.81(2H, d, *J*= 8Hz, H-2 and H-6).

Ethyl 4-acetoxybenzoate (**2**). Colourless viscous mass (lit[12] mp 34°C); UV(MeOH): 233 nm; ^{1}H NMR($CDCl_3$): δ 1.30(3H, t, *J*=7Hz, OCH_2CH_3), 2.24(3H, s, $OCOCH_3$), 4.14(2H, q, *J*=7Hz, OCH_2CH_3), 6.80(2H, d, *J*=8Hz, H-3 and H-5), 7.64(2H, d, *J*=8Hz, H-2 and H-6).

Methyl 2-acetoxybenzoate (**3**). Colourless viscous mass (lit[9a] mp 49°C); UV(MeOH): 236, 275 nm; ^{1}H NMR($CDCl_3$): δ 2.30(3H, s, $OCOCH_3$), 3.74(3H, s, $COOCH_3$), 6.74-7.83(4H, m, H-3, H-4, H-5 and H-6).

Ethyl 2-acetoxybenzoate (**4**). Colourless oil (lit[9a] bp 272°C); UV(MeOH): 230, 275 nm; ^{1}H NMR($CDCl_3$): δ 1.32(3H, t, *J*=7Hz, OCH_2CH_3), 2.30(3H, s, $OCOCH_3$), 4.32(2H, q, *J*=7Hz, OCH_2CH_3), 7.06-8.10(4H, m, H-3, H-4, H-5 and H-6).

Methyl 2,4-diacetoxybenzoate (**5**).[17] White solid, mp 50-51°C (lit[10,17] mp 62-63°C); IR(Nujol): 1780, 1720, 1620, 1380, 1280, 1260, 1200 cm^{-1}; UV (MeOH): 209, 227, 284 nm; ^{1}H NMR ($CDCl_3$): δ 2.29 (3H, s, $OCOCH_3$), 2.33 (3H, s, $OCOCH_3$), 3.85(3H, s, $COOCH_3$), 6.94(1H, d, *J*=2Hz, H-3), 7.06(1H, dd, *J*=2 & 8Hz, H-5), 8.02 (1H, d, *J*=8Hz, H-6); ^{13}C NMR ($CDCl_3$): δ 20.80($OCOCH_3$), 21.00($OCOCH_3$), 52.10($COOCH_3$), 117.22(C-3), 119.03(C-5), 120.43(C-1), 132.59(C-6), 151.53(C-2), 154.37(C-4), 164.09, 168.16 and 169.19 (3xC=O); EIMS, *m/z* (%): 252([M]$^+$, 15), 221([M-OCH_3]$^+$, 48), 210([M-CH_2CO]$^+$, 40), 168([M-2xCH_2CO]$^+$, 100), 136([M-2xCH_2CO-CH_3OH]$^+$, 92), 108(8), 57(6), 43(32).

Ethyl 2,4-diacetoxybenzoate (**6**). Colourless oil; IR (Nujol) : 1780, 1730, 1620, 1380, 1285, 1260, 1200 cm^{-1}; UV (MeOH): 209, 236, 295nm; ^{1}H NMR ($CDCl_3$): δ1.37 (3H, t, *J*=7Hz,OCH_2CH_3), 2.30 (3H, s, $OCOCH_3$), 2.36(3H, s, $OCOCH_3$), 4.35 (2H, q, *J*=7Hz, OCH_2CH_3), 6.93 (1H, d, *J*=2.3Hz, H-3), 7.14 (1H, dd, *J*=2.3 & 8.7Hz, H-5), 8.09(1H, d, *J*=8.7Hz, H-6); ^{13}C NMR ($CDCl_3$): δ 14.06(OCH_2CH_3), 20.80($OCOCH_3$), 20.94($OCOCH_3$), 61.10($COOCH_2CH_3$), 117.13(C-3), 119.03(C-5), 120.8(C-1), 132.57(C-6), 151.32(C-2), 154.24(C-4), 163.79, 168.26 and 169.29(3xC=O); EIMS, *m/z*(%): 266([M]$^+$, 5), 224([M-CH_2CO]$^+$, 22), 221([M-OC_2H_5]$^+$, 5), 182([M-2xCH_2CO]$^+$, 86), 137([M-2xCH_2CO-OC_2H_5]$^+$, 30), 136([M-2xCH_2CO-C_2H_5OH]$^+$, 100), 108(12), 69(3), 43(20).

Methyl 3,5-diacetoxybenzoate (**7**). White solid, mp 59-60°C; IR(Nujol): 1780, 1730, 1600, 1370, 1315, 1195 cm^{-1}; UV(MeOH): 209, 227, 284nm; ^{1}H NMR ($CDCl_3$): δ 2.29(6H, s, 2x$OCOCH_3$), 3.90(3H, s, $COOCH_3$), 7.13(1H, t, *J*=2.2Hz, H-4), 7.65 (2H, d, *J*=2.2Hz, H-2 and H-6); ^{13}C NMR($CDCl_3$): δ20.89(2x$OCOCH_3$), 52.39($COOCH_3$), 119.99(C-4), 120.18(C-2 and C-6), 132.12(C-1), 158.87(C-3 and C-5), 165.24, 168.65(3xC=O); EIMS, *m/z*(%): 252 ([M]$^+$, 60), 221([M-OCH_3]$^+$, 9), 210([M-CH_2CO]$^+$, 34), 168([M-2xCH_2CO]$^+$, 100), 137([M-2xCH_2CO-OCH_3]$^+$, 35), 109(5), 57(6), 43(22).

Ethyl 3,5-diacetoxybenzoate(**8**). White solid, mp 49-50°C; IR (Nujol) : 1780, 1730, 1600, 1370, 1315, 1195 cm^{-1}; UV(MeOH): 209, 227, 284nm; ^{1}H NMR($CDCl_3$) : δ1.37 (3H, t, *J*=7Hz, OCH_2CH_3), 2.30 (6H, s,

2xOCOCH$_3$), 4.32 (2H, q, *J*=7Hz, OC*H*$_2$CH$_3$), 7.13(1H, t, *J*=2.2Hz, H-4), 7.65(2H, d, *J*=2.2Hz, H-2 and H-6); ^{13}C NMR (CDCl$_3$): δ14.21(COOCH$_2$*C*H$_3$), 20.97(2xOCO*C*H$_3$), 61.48(O*C*H$_2$CH$_3$), 119.96(C-4), 120.20(C-2 and C-6), 132.58(C-1), 158.88(C-3 and C-5), 164.79 and 168.74(3xC=O); EIMS, *m/z* (%): 266 ([M]$^+$, 6), 224([M-CH$_2$CO]$^+$, 25), 221([M-OC$_2$H$_5$]$^+$, 10), 182([M-2xCH$_2$CO]$^+$, 100), 137([M-2xCH$_2$CO-OC$_2$H$_5$]$^+$, 22), 110(10), 71(5), 43(20).

Methyl 2,3,4-triacetoxybenzoate (**9**). White solid, mp 93-95°C; IR (Nujol) : 1790, 1770, 1725, 1620, 1350, 1300, 1195, 1160 cm^{-1}; UV(MeOH): 209, 231, 275nm; ^{1}H NMR (CDCl$_3$): δ 2.26(3H, s, OCOCH$_3$), 2.29(3H, s, OCOCH$_3$), 2.32(3H, s, OCOCH$_3$), 3.84(3H, s, COOCH$_3$), 7.19(1H, d, *J*=8.8Hz, H-5), 7.90(1H, d, *J*=8.8Hz, H-6); ^{13}C NMR (CDCl$_3$): δ 19.90(OCO*C*H$_3$), 20.38(OCO*C*H$_3$), 20.49(OCO*C*H$_3$), 52.20(COO*C*H$_3$), 120.21(C-5), 121.77(C-1), 128.50(C-6), 135.84(C-3), 144.28(C-4), 146.82(C-2), 163.68, 166.80, 167.10 and 167.93(4xC=O); EIMS, *m/z* (%): 310([M]$^+$, 2), 279([M-OCH$_3$]$^+$, 4), 268([M-CH$_2$CO]$^+$, 12), 226([M-2xCH$_2$CO]$^+$, 55), 184([M-3xCH$_2$CO]$^+$, 93), 152([M-3xCH$_2$CO - CH$_3$OH]$^+$, 100), 138 (9), 60(7), 43(35).

Ethyl 2,3,4-triacetoxybenzoate (**10**). White solid, mp 71°C; IR (Nujol) : 1775, 1730, 1620, 1380, 1280, 1190, 1160 cm^{-1}; UV(MeOH): 209, 231, 272nm; ^{1}H NMR (CDCl$_3$): δ1.32 (3H, t, *J*=7.1Hz, OCH$_2$C*H*$_3$), 2.28 (3H, s, OCOCH$_3$), 2.30(3H, s, OCOCH$_3$), 2.33(3H, s, OCOCH$_3$), 4.27(2H, q, *J*=7.1Hz, OC*H*$_2$CH$_3$), 7.20(1H, d, *J*=8.9Hz, H-5), 7.92(1H, d, *J*=8.9Hz,H-6); ^{13}C NMR (CDCl$_3$): δ 14.12(OCH$_2$*C*H$_3$), 20.01(OCO*C*H$_3$), 20.51(OCO*C*H$_3$), 20.61(OCO*C*H$_3$), 61.29(O*C*H$_2$CH$_3$), 120.24(C-5), 122.21(C-1), 128.61(C-6), 135.78(C-3), 144.11(C-4), 146.71(C-2), 163.32, 166.88, 167.17 and 167.93(4xC=O); EIMS, *m/z* (%): 324([M]$^+$, 3), 282([M-CH$_2$CO]$^+$, 5), 240([M-2xCH$_2$CO]$^+$, 36), 237([M-CH$_2$CO - OC$_2$H$_5$]$^+$, 3), 198([M-3xCH$_2$CO]$^+$, 78), 153([M-3xCH$_2$CO - OC$_2$H$_5$])$^+$, 15), 152([M-3xCH$_2$CO - C$_2$H$_5$OH]$^+$, 100), 138(8), 60(9), 43(32).

Methyl 3,4,5-triacetoxybenzoate (**11**). White solid, mp 122-24°C (lit[9b,13] mp 126-28°C and 120-22°C); UV (MeOH): 209, 231, 275 nm; ^{1}H NMR(CDCl$_3$): δ 2.25(9H, s, 3xOCOCH$_3$), 3.90(3H, s, COOCH$_3$), 7.92(2H, s, H-2 and H-6).

Ethyl 3,4,5-triacetoxybenzoate (**12**). White solid, mp 138°C (lit[9b] mp 138°C); IR(Nujol): 1780, 1680, 1622, 1480, 1420, 1260, 1180 cm^{-1}; UV(MeOH): 206, 233nm; ^{1}H NMR (CDCl$_3$) : δ1.33 (3H, t, *J*=7.1Hz,OCH$_2$C*H*$_3$), 2.28 (9H, s, 3xOCOCH$_3$), 4.31(2H, q, *J*=7.1Hz, OC*H*$_2$CH$_3$), 7.79 (2H, s, H-2 and H-6); ^{13}C NMR(CDCl$_3$): δ 13.95(OCH$_2$*C*H$_3$), 19.80(2xOCO*C*H$_3$), 20.21(OCO*C*H$_3$), 61.33(O*C*H$_2$CH$_3$), 121.86(C-2 and C-6), 128.39(C-1), 138.33(C-4), 143.18(C-3 and C-5), 164.12, 166.17, 167.37(4x>C=O); EIMS, *m/z* (%): 324([M]$^+$, 8), 282([M-CH$_2$CO]$^+$, 12), 240([M-2xCH$_2$CO]$^+$, 56), 237([M-CH$_2$CO - OC$_2$H$_5$]$^+$, 6), 198([M-3xCH$_2$CO]$^+$, 100), 153([M-3xCH$_2$CO - OC$_2$H$_5$]$^+$, 12), 138(6), 60(10), 43(33).

2,4-Diacetoxyacetophenone (**13**). Viscous mass (lit[9g] mp 38°C); UV(MeOH): 245, 281(sh) nm; ^{1}H NMR (CDCl$_3$): δ 2.22(3H, s, OCOCH$_3$), 2.26(3H, s, OCOCH$_3$), 2.40(3H, s, COCH$_3$), 6.74-6.92 (2H, m, H-3 and H-5), 7.50(1H, d, *J*=8Hz, H-6).

2,4-Diacetoxybenzophenone (**14**). White solid, mp 77-78°C (lit[9h] mp 78°C); UV(MeOH): 224, 254, 326nm;

^{1}H NMR($CDCl_3$): δ 1.85(3H, s, $OCOCH_3$), 2.22(3H, s, $OCOCH_3$), 6.71(2H, m, H-3 and H-5), 7.28(6H, m, H-6 and CO*Ph*).

Methyl 4-hydroxybenzoate (**15**). White solid, mp 127°C (lit[9c,14] mp 131°C and 127-29°C); UV(MeOH): 210, 255 nm; + NaOAc: 210, 255 nm; + NaOMe: 210, 295 nm; ^{1}H NMR($CDCl_3$): δ 3.75(3H, s, $COOCH_3$), 4.92(1H, brs, OH), 6.55(2H, d, *J*=8Hz, H-3 and H-5), 7.62(2H, d, *J*=8Hz, H-2 and H-6).

Ethyl 4-hydroxybenzoate (**16**). White solid, mp 116°C;(lit[9c,14] mp 112°C and 116°C); UV(MeOH): 210, 255 nm; + NaOAc: 210, 255 nm; + NaOMe: 210, 295 nm; ^{1}H NMR($CDCl_3$): δ 1.35(3H, t, *J*=7Hz, CH_2CH_3), 4.35(2H, q, *J*=7Hz, -OCH_2CH_3), 6.95(2H, d, *J*=8Hz, H-3 and H-5), 7.99(2H, d, *J*=8Hz, H-2 and H-6).

M*ethyl 2-hydroxybenzoate* (**17**). Colourless oil (lit[9d] bp 223°C); UV(MeOH): 210, 235, 304 nm; + $AlCl_3$: 210, 250, 345nm; + NaOMe: 245, 340 nm; ^{1}H NMR(CCl_4): δ 3.82(3H, s, $COOCH_3$), 6.80-7.84(4H, m, H-3, H-4, H-5 and H-6), 10.40(1H, s, chelated OH).

Ethyl 2-hydroxybenzoate (**18**). Colourless oil (lit[9d] bp 231-35°C); UV(MeOH): 210, 235,305 nm; + $AlCl_3$: 210, 250, 350nm; + NaOMe: 245, 337 nm; ^{1}H NMR(CCl_4): δ 1.35(3H, t, *J*=7Hz, OCH_2CH_3), 4.34(2H, q, *J*=7Hz, OCH_2CH_3), 6.71-7.90(4H, m, H-3, H-4, H-5 and H-6), 10.8(1H, s, chelated OH).

Methyl 2-acetoxy-4-hydroxybenzoate (**19**). White solid, mp 138°C; IR (KBr) : 3350, 1720, 1620, 1595, 1382, 1280, 1260, 1230 cm^{-1}; UV (MeOH): 212, 254nm; + $AlCl_3$: 214, 252nm; +NaOAc : 214, 254nm; +NaOMe : 220, 294nm; ^{1}H NMR (DMSO-d_6): δ2.25 (3H, s, $OCOCH_3$), 3.74 (3H, s, $COOCH_3$), 6.54 (1H, d, *J*=2.4Hz, H-3), 6.75 (1H, dd, *J*= 2.4 and 8.7Hz, H-5), 8.00 (1H, d, *J*=8.7Hz, H-6); ^{13}C NMR (DMSO-d_6): δ20.67 ($OCOCH_3$), 52.20 ($COOCH_3$), 110.49 (C-3), 113.09 (C-5), 113.15 (C-1), 132.92 (C-6), 152.14 (C-2), 162.68 (C-4), 164.03 , 168.89 (2xC=O); EIMS, *m/z* (%) : 210 ([M]$^+$, 10), 179 ([M-OCH_3]$^+$, 4), 168 ([M-CH_2CO]$^+$, 80), 136 ([M-CH_2CO - CH_3OH]$^+$,100), 108 (19), 43 (15).

Methyl 2,4-dihydroxybenzoate (**20**). White solid, mp 72°C (lit[9c,14] mp 78-80°C and 118-19°C); UV(MeOH): 209, 259, 296 nm; + $AlCl_3$: 210, 276, 323 nm; + NaOAc: 209, 259, 296nm; + NaOMe: 209, 236, 304 nm; ^{1}H NMR (Acetone-d_6): 3.90(3H, s, $COOCH_3$), 6.42-6.68(2H, m, H-3 and H-5), 7.80(1H, d, *J*=8Hz, H-6), 8.90(1H, brs, chelated OH).

Ethyl 2-acetoxy-4-hydroxybenzoate (**21**). White solid, mp 131°C; IR (KBr) : 3320, 1745, 1620, 1370, 1336, 1240, 1250 cm^{-1}; UV (MeOH): 214, 252nm; +$AlCl_3$: 214, 252nm; +NaOAc : 212, 254nm; +NaOMe : 212, 292nm; ^{1}H NMR (DMSO-d_6) : δ1.25 (3H, t, *J*=7.1Hz, OCH_2CH_3), 2.24 (3H, s, $OCOCH_3$), 3.30 (1H, brs, Ar-OH), 4.20 (2H, q, *J*=7.1Hz, OCH_2CH_3), 6.53 (1H, d, *J*=2.4Hz, H-3), 6.76 (1H, dd, *J*=2.4 and 8.7Hz, H-5), 7.81 (1H, d, *J*=8.7Hz, H-6); ^{13}C NMR (DMSO-d_6): δ14.04 (OCH_2CH_3), 20.72 ($OCOCH_3$), 60.11 (OCH_2CH_3), 110.44 (C-3), 113.14 (C-5), 113.49 (C-1), 132.94 (C-6), 152.00 (C-2), 162.55 (C-4), 163.66 and 168.81 (2xC=O); EIMS, *m/z* (%) : 224 ([M]$^+$, 8), 182 ([M-CH_2CO]$^+$, 62), 179 ([M-OC_2H_5],$^+$ 4), 136 ([M-CH_2CO - C_2H_5OH]$^+$, 100), 108 (17), 44 (12), 43 (11).

Ethyl 2,4-dihydroxybenzoate (**22**). White solid, mp 71-72°C (lit[11] mp 69-70°C); UV(MeOH): 209, 259, 294 nm; +$AlCl_3$: 209, 278, 323nm; + NaOAc: 209, 259, 294 nm; + NaOMe 209, 236, 302 nm; ^{1}H NMR(Acetone-d_6): δ 1.35(3H, t, *J*=7Hz, OCH_2CH_3), 4.35(2H, q, *J*=7Hz, OCH_2CH_3), 6.35-6.56 (2H, m, H-3 and H-5), 7.22(1H, brs, OH), 7.80(1H, d, *J*=8Hz, H-6), 10.8(1H, brs, chelated OH).

Methyl 3-acetoxy-5-hydroxybenzoate (**23**). White solid, mp 118-19°C (lit[15] mp 120-21°C and 118-21°C); IR (KBr) : 3350, 1770, 1705, 1608, 1380, 1340, 1200 cm^{-1}; UV (MeOH): 210, 300nm; +NaOAc : 210, 300nm; +NaOMe : 275, 340nm; ^{1}H NMR (CD_3OD): δ 2.35 (3H, s, $OCOCH_3$), 3.96 (3H, s, $COOCH_3$), 6.85 (1H, t, *J*=2.3Hz, H-4), 7.28(1H, t, *J*=2.3Hz, H-6), 7.38 (1H, t, *J*=2.3Hz, H-2); ^{13}C NMR (CD_3OD): δ 21.15($OCOCH_3$), 53.07($COOCH_3$), 115.01(C-4), 115.07(C-2 and C-6), 133.50(C-1), 153.40(C-3), 160.08(C-5), 167.99, 171.14(2xC=O); EIMS, *m/z*(%): 210([M]$^+$, 15), 179([M-OCH_3]$^+$, 13), 168([M-CH_2CO]$^+$, 100), 137([M-CH_2CO-OCH_3]$^+$, 77), 109(16), 43(20).

Methyl 3,5-dihydroxybenzoate (**24**). White solid, mp 163-64°C (lit[11] mp 163-65°C); UV(MeOH): 210, 250, 305 nm; + NaOAc: 250, 305nm; + NaOMe: 280, 345nm; ^{1}H NMR(Acetone-d_6): δ 3.85(3H, s, $COOCH_3$), 6.72(1H, s, H-4), 7.20(2H, s, H-2 and H-6), 8.5(2H, brs, 2xOH).

Ethyl 3-acetoxy-5-hydroxybenzoate (**25**). White solid, mp 108-10°C; IR (Nujol) : 3390, 1740, 1720, 1618, 1380, 322, 1140, 1150 cm^{-1}; UV (MeOH) : 210, 300nm; +NaOAc; 240, 300nm; +NaOMe : 275, 340nm; ^{1}H NMR (CD_3OD): δ1.43 (3H, t, *J*=7.1Hz, OCH_2CH_3), 2.36 (3H, s, $OCOCH_3$), 4.38 (2H, q, *J*=7.1Hz, OCH_2CH_3), 6.84 (1H, t, *J*=2.2Hz, H-4), 7.27(1H, dd, *J*=1.4 and 2Hz, H-6), 7.39(1H, dd, *J*=1.4 and 2.3Hz, H-2); ^{13}C NMR(CD_3OD): δ 14.83(OCH_2CH_3), 21.15($OCOCH_3$), 62.65(OCH_2CH_3), 115.00(C-2, C-4 and C-6), 133.85(C-1), 153.40(C-3), 160.09(C-5), 167.51 and 171.16(2xC=O); EIMS, *m/z*(%): 224([M]$^+$, 18), 182([M-CH_2CO]$^+$, 100), 179([M-OC_2H_5]$^+$, 16), 137([M-CH_2CO - OC_2H_5]$^+$, 62), 110(20), 43(22).

Ethyl 3,5-dihydroxybenzoate(**26**). White solid, mp 115-22°C (lit[11,14] mp 127-28°C); UV(MeOH): 210, 250, 305nm; +NaOAc: 210, 305nm; +NaOMe: 230, 345nm; ^{1}H NMR($CDCl_3$): δ 1.25(3H, t, *J*=7Hz, OCH_2CH_3), 4.25(2H, q, *J*=7Hz, OCH_2CH_3), 6.65(1H, s, H-4), 7.14(2H, s, H-2 and H-6), 8.46(2H, brs, 2xOH).

Methyl 3,4-diacetoxy-2-hydroxybenzoate (**27**). White solid, mp 95-96°C; IR (Nujol): 3200(br), 1780, 1680, 1620, 1540, 1520, 1380, 1260, 1200, 1140, 1060 cm^{-1}; UV (MeOH): 210, 248, 297nm; +$AlCl_3$+ HCl: 210, 248, 297, 326nm; +NaOAc: 210, 258, 296nm; +NaOMe: 210, 245, 296, 410nm; ^{1}H NMR(DMSO-d_6): δ 2.26 and 2.28 (6H, 2s, 2x$OCOCH_3$), 3.75(3H, s, $COOCH_3$), 6.90 (1H, d, *J*=8.8Hz, H-5), 7.72 (1H, d, *J*=8.8Hz, H-6), 10.76 (1H, s, chelated OH); ^{13}C NMR(DMSO-d_6): δ 19.89($OCOCH_3$), 20.17($OCOCH_3$), 52.20($COOCH_3$), 108.17(C-1 and C-5), 127.56(C-6), 154.77(C-3), 155.70(C-4), 163.76(C-2), 167.67(C=O), 167.98(C=O), 169.39(C=O); EIMS, *m/z* (%) : 268 ([M]$^+$, 2), 237 ([M-OCH_3]$^+$, 1), 226 ([M-CH_2CO]$^+$, 17), 184 ([M-2xCH_2CO]$^+$, 52), 152 ([M-2xCH_2CO - CH_3OH]$^+$, 100), 124 (8), 43 (15).

Methyl 2,3,4-trihydroxybenzoate (**28**). White solid, mp 150°C (lit[9f] mp 150°C); UV(MeOH): 209, 227, 264 nm; + $AlCl_3$:220, 234, 290nm; + NaOAc: 209, 227, 264nm; + NaOMe: 209, 278, 359nm; ^{1}H NMR (CD_3OD):

δ3.80(3H, s, $COOCH_3$), 6.42(1H, d, *J*=8Hz, H-5), 7.22(1H, d, *J*=8Hz, H-6).

Ethyl 3,4-diacetoxy-2-hydroxybenzoate (**29**). White solid, mp 95°C; IR(KBr): 3100, 1780, 1680, 1620, 1480, 1420, 1270, 1200 cm^{-1}; UV(MeOH): 210, 246, 297nm; +$AlCl_3$:226, 256, 317nm; +NaOAc: 210, 260, 294nm; NaOMe: 209, 232, 284, 360nm; ^1H NMR(CD_3OD): δ 1.44(3H, t, *J*=7.1Hz, OCH_2CH_3), 2.36(3H, s, $OCOCH_3$), 2.38(3H, s, $OCOCH_3$), 4.49(2H, q, *J*=7.1Hz, OCH_2CH_3), 6.87(1H, d, *J*=8.9Hz, H-5), 7.87(1H, d, *J*=8.9Hz, H-6); ^{13}C NMR (CD_3OD): δ 14.66(OCH_2CH_3), 20.27($OCOCH_3$), 20.70($OCOCH_3$), 63.38(OCH_2CH_3), 114.81(C-1), 115.0 (C-5), 128.57(C-6), 149.79(C-3 and C-4), 157.30(C-2), 169.39, 169.54 (3xC=O); EIMS, *m/z*(%): 282($[M]^+$, 1), 240($[M-CH_2CO]^+$, 18), 237($[M-OC_2H_5]^+$,1), 198($[M-2xCH_2CO]^+$, 58), 152($[M-2xCH_2CO-C_2H_5OH]^+$, 100), 124(5), 43(16).

Ethyl 2,3,4-trihydroxybenzoate (**30**). White solid, mp 82-83°C (lit[9f, 14] mp 102°C); UV(MeOH): 212, 269 nm; + $AlCl_3$: 227, 293nm; + NaOAc: 224, 273 nm; + NaOMe: 230, 314 nm; ^1H NMR($CDCl_3$+CD_3OD): δ 1.50(3H, t, *J*=7Hz, OCH_2CH_3), 4.40(2H, q, *J*=7Hz, OCH_2CH_3), 6.60(1H, d, *J*=8Hz, H-5), 7.40(1H, d, *J*=8Hz, H-6).

Methyl 3,4,5-trihydroxybenzoate (**32**). White solid, mp 157°C (lit[9b] mp 157°C);. UV(MeOH): 218, 272 nm; + NaOAc: 218, 272 nm; + NaOAc + H_3BO_3: 221, 291 nm; + NaOMe: 215, 297 nm; ^1H NMR(DMSO-d_6): δ 3.81(3H, s, $COOCH_3$), 7.0(2H, s, H-2 and H-6), 8.93(3H, brs, 3xOH).

Ethyl 3,4,5-trihydroxybenzoate (**34**). White solid, mp 162°C (lit[9b] mp 160-62°C). UV(MeOH): 221, 272 nm; + NaOAc: 221, 272 nm; + NaOAc + H_3BO_3: 222, 296 nm; + NaOMe: 218, 236(sh), 284, 316 nm; ^1H NMR(DMSO- d_6): δ 1.32(3H, t, *J*=7Hz, OCH_2CH_3), 4.21(2H, q, *J*=7Hz, OCH_2CH_3), 6.95(2H, s, H-2 and H-6), 7.98(3H, brs, 3xOH).

2,4-Dihydroxyacetophenone (**36**). White solid, mp 147°C (lit[9g] mp 147°C); UV(MeOH): 230, 272, 321 nm; + $AlCl_3$: 227, 300, 350(sh); + NaOAc: 230, 272, 320 nm; + NaOMe: 224, 248nm; ^1H NMR (Acetone-d_6): δ 2.41(3H, s, $COCH_3$), 6.32(2H, m, H-3 and H-5), 7.25(1H, d, *J*=8Hz, H-6), 12.00(1H, s, chelated OH).

2,4-Dihydroxybenzophenone (**38**). White solid, mp 143°C (lit[9h] mp 142.6-144.6°C); UV(MeOH): 224, 242, 290, 320 nm; +$AlCl_3$: 224, 239, 317, 380 nm; + NaOAc: 224, 242, 290, 326 nm; + NaOMe: 224, 244 nm; ^1H NMR (Acetone-d_6): δ 6.0(2H, m, H-3 and H-5), 7.1(6H,m, H-6 and CO*Ph*), 12.0(1H, s, chelated OH).

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14. Some compounds, in particular the hydroxy esters, are expected to have two melting points as they can partly crystallize with water of crystallization; the two melting points correspond to the anhydrous form and the hydrated form. In the literature, some compounds of this type are reported to have two melting points. In cases where the melting point of our compound does not correspond to that reported in literature for the same compound, we believe our compound exists in a form different than the one for which the melting point is given.
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17. The compound **5** was reported in the year 1952,[10] no spectral data was published. It is reported to melt at 62-63°C, our sample of **5**, however has been fully characterised from its different spectral data (IR, UV, ^{1}H and ^{13}C NMR, and mass) and it melted at 50-51°C.

18. Physical and spectral data of the two inseparable mixtures obtained in the lipase-mediated deacetylation reactions on triacetoxybenzoates **11** and **12**. *Methyl 3,5-diacetoxy-4-hydroxybenzoate* (**31**) + *Methyl 3,4,5-triacetoxybenzoate* (**11**): White solid, mp 108-09°C; IR (KBr) : 3180, 1780, 1720, 1610, 1380, 1330, 1240, 1250, 1200, 1180 cm^{-1}; UV (MeOH): 210, 230, 280nm; +NaOAc : 210, 235, 280nm; +NaOMe : 210, 294nm; ^1H NMR (DMSO-d_6): δ2.29 and 2.33(2s, integrating in the ratio 2:1, $OCOCH_3$), 3.87 (s, $COOCH_3$), 7.79 (s, H-2 and H-6); ^{13}C NMR (DMSO-d_6): δ19.82 ($OCOCH_3$), 20.35($OCOCH_3$), 52.67 ($COOCH_3$), 121.90 (C-2 and C-6), 127.57 (C-1), 143.34 (C-3 and C-5), 164.37, 166.20 and 167.30 (C=O); EIMS, *m/z* (%): 310(2), 268 (12), 237 ([M-OCH_3]$^+$, 5), 226 ([M-CH_2CO]$^+$, 46), 184 ([M-2xCH_2CO]$^+$, 100), 153 ([M-2xCH_2CO - OCH_3]$^+$, 36), 125 (5), 43 (35). *Ethyl 3,5-diacetoxy-4-hydroxybenzoate* (**33**) + *Ethyl 3,4,5-triacetoxybenzoate* (**12**). White solid, mp 140°C; IR (KBr) : 3180, 1780, 1720, 1620, 1380, 1330, 1200, 1160 cm^{-1}; UV (MeOH): 210, 230, 280nm; +NaOAc : 210, 235, 280nm; +NaOMe: 215, 230, 302nm; ^1H NMR (DMSO-d_6): δ1.32 (t, J=7.1Hz, OCH_2CH_3), 2.30 and 2.33 (2s, integrating in the ratio 2:1, $OCOCH_3$), 4.34 (q, J=7.1Hz, OCH_2CH_3), 7.78 (s, H-2 and H-6); ^{13}C NMR (DMSO-d_6): δ13.99 (-OCH_2CH_3), 19.75 ($OCOCH_3$), 20.28($OCOCH_3$), 61.40 (OCH_2CH_3), 121.77 (C-2 and C-6), 127.83 (C-1), 143.30 (C-3 and C-5), 163.81, 166.80 and 167.92 (C=O); EIMS, *m/z* (%) : 324(1), 282 (6), 240 ([M-CH_2CO]$^+$, 36), 237 ([M-OC_2H_5]$^+$, 5), 198 ([M-2xCH_2CO]$^+$, 100), 183 (6), 170 (20), 153 ([M-2xCH_2CO-OC_2H_5]$^+$, 42), 125 (5).

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