

signal at m/z 466 (see Supporting Information; Figure S3). The observed mass and isotope patterns are consistent with the ion $[\text{Zn}(\text{QH})\{\text{MeIm}(\text{Py})_2\}]^+$, which is the protonated form of the reduced species of $[\text{Zn}(\text{Q}^{\cdot-})\{\text{MeIm}(\text{Py})_2\}]^+$.

In conclusion, the Zn^{II} ion in the SOD model complex has been shown to play the essential role in facilitating the reduction of $\text{Q}^{\cdot-}$ by coordination of $\text{Q}^{\cdot-}$ to the Zn^{II} ion. The oxidation of $\text{Q}^{\cdot-}$ is also facilitated by the Zn^{II} ion, since the reduction potential of the Cu^{II} center in the imidazolate-bridged $\text{Cu}^{\text{II}}-\text{Zn}^{\text{II}}$ heterodinuclear complex **1** is shifted to a more positive value (0.21 V) relative to that without a Zn^{II} ion.^[10] Thus, the Zn^{II} ion can facilitate both the oxidation and reduction of $\text{Q}^{\cdot-}$. Essentially the same mechanism may also be applied to the disproportionation of $\text{O}_2^{\cdot-}$ catalyzed by Zn,Cu-SOD .

Experimental Section

To a deaerated solution (25 mL) of *p*-benzoquinone (8.10 mg) in EtCN and hydroquinone (8.25 mg) was added two equivalents of 1M $\text{Bu}_4\text{NOH} \cdot \text{MeOH}$ solution (72 μL) to make the stock solution of *p*-benzosemiquinone radical anion $\text{Q}^{\cdot-}$ ($6.0 \times 10^{-3} \text{ M}$). The reactions of imidazolate-bridged $\text{Cu}^{\text{II}}-\text{Zn}^{\text{II}}$ heterodinuclear and $\text{Cu}^{\text{II}}-\text{Cu}^{\text{II}}$ homodinuclear SOD model complexes and semiquinone radical anion were performed in a UV/Vis cell (path length 1 cm) which was held in a Unisoku temperature-controlled ($\pm 0.5^\circ\text{C}$) cell holder designed for low-temperature experiments. After the deaerated solution of the SOD model complexes ($0.1 \times 10^{-4} \text{ M}$) in the cell had been kept at the desired temperature for several minutes, semiquinone radical anion was added by syringe. Formation of the $\text{Cu}^{\text{I}}-\text{Q}$ complexes was followed by monitoring the absorption change at 585 nm. The rate constant k_{obs} for the stoichiometric disproportionation of $\text{Q}^{\cdot-}$ was determined by monitoring the decrease in the absorption band due to $\text{Q}^{\cdot-}$ ($\lambda_{\text{max}} = 422 \text{ nm}$).

Frozen-solution ESR spectra were recorded on a JEOL JES-RE1X X-band spectrometer equipped with a standard low-temperature apparatus. All spectra were recorded at 77 K in quartz tubes with 4 mm inner diameters. The g values were calibrated with a Mn^{II} marker as a reference.

Resonance Raman spectra were excited at 632.8 nm with an He–Ne laser and detected with a JASCO NR-1800 triple polychromator equipped with a liquid-nitrogen-cooled Princeton Instruments CCD detector. Raman measurements were carried out with a spinning cell, and the laser power was adjusted to 50 mW at the sample point. Raman shifts were calibrated with acetonitrile; the accuracy of the peak positions of the Raman bands was $\pm 1 \text{ cm}^{-1}$.

ESI mass spectra were obtained with an API 150 triple quadrupole mass spectrometer (PE-Sciex) in positive-ion detection mode, equipped with an ion-spray interface. The sprayer was held at a potential of 5.0 kV, and compressed N_2 was employed to assist liquid nebulization. The positive-ion ESI mass spectra were measured in the range m/z 100–1000.

Received: August 22, 2000 [Z15682]

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New Family of Cyclopropanating Reagents: Synthesis, Reactivity, and Stability Studies of Iodomethylzinc Phenoxides**

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The cyclopropanation of olefins is a very useful process in synthetic organic chemistry. Cyclopropane moieties are found in many natural^[1] and unnatural products^[2] possessing interesting biological activities.^[3] These units are also very useful synthons for further synthetic transformations.^[4]

Amongst the different methods of cyclopropanation, the Simmons–Smith reaction^[5] has stimulated a considerable

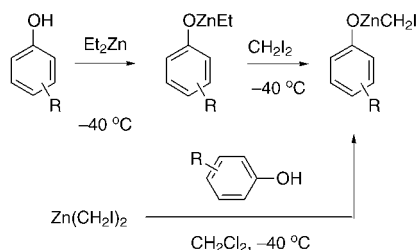
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[**] This work was supported by the E.W.R. Steacie Fund, the National Science and Engineering Research Council (NSERC) of Canada, Merck Frosst Canada, Boehringer Ingelheim (Canada), F.C.A.R. (Québec), and the Université de Montréal. J.M. and S.F. are grateful to NSERC and F.C.A.R. respectively for postgraduate fellowships.

amount of interest within the chemical community.^[6] Although the initial method for the preparation of zinc carbenoids with a zinc/copper couple was cumbersome, several synthetically more accessible and reproducible methods quickly followed.^[7] Notable amongst these is the protocol of Furukawa et al. which prepares a zinc carbenoid from Et₂Zn and CH₂I₂.^[8] This landmark observation allowed for the reaction to take place in noncomplexing solvents, such as dichloromethane or 1,2-dichloroethane,^[9] which greatly improved the reactivity of zinc carbenoids for the cyclopropanation of olefins.

Despite the fact that zinc carbenoids have been studied quite extensively, very little work has been done to modify the nature of the R group of the zinc reagent “RZnCH₂X” and to compare the reactivity of the new reagents to the classical ones (when R = I, Et, CH₂X). A few years ago, we demonstrated that a new class of reagents “ROZnCH₂I” (R = alkyl or allyl) are unreactive towards olefins in the absence of a Lewis acid.^[10] Recently, Shi and co-workers reported^[11] that a reagent prepared by mixing stoichiometric quantities of Et₂Zn, trifluoroacetic acid, and CH₂I₂ (which presumably form “RCOOZnCH₂I”) displayed an increased reactivity towards stilbene, a substrate that is often unreactive using previously reported Simmons–Smith protocols. In this communication, we report that some reagents of general structure “ArOZnCH₂I” are very reactive species for the cyclopropanation of unfunctionalized olefins. This synthetically useful observation is a first step towards the development of an enantioselective cyclopropanation method for unfunctionalized olefins.

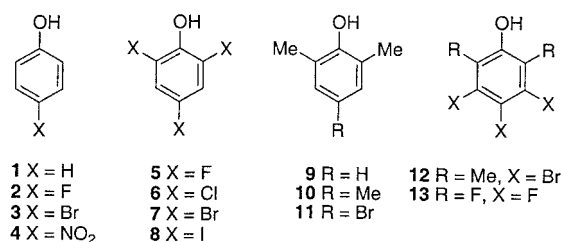
The carbenoids ArOZnCH₂I can be easily prepared by two different methods (Scheme 1). The phenol can be deprotonated upon treatment with Et₂Zn, and a subsequent metal–halogen exchange process with CH₂I₂ leads to the formation



Scheme 1. Two methods for the synthesis of ArOZnCH₂I from the corresponding phenol.

of the cyclopropanating reagent. Alternatively, one equivalent of the phenol can be treated with one equivalent of Zn(CH₂I)₂.^[8] The first method is generally preferred since it uses only one equivalent of diiodomethane and it does not necessitate the prior formation of the less stable bis(iodomethyl)zinc.

Several phenols (**1**–**13**) possessing different substitution patterns and acidities were screened (Scheme 2). To test the reactivity of these reagents, three relatively unreactive olefins were selected: α -methylstyrene (**14**), styrene (**15**), and indene (**16**). These unfunctionalized olefins were treated with the zinc phenoxide reagents and the results are shown in Table 1.^[12]



Scheme 2. Phenolic precursors for the cyclopropanating reagents.

Table 1. Reactivity of ArOZnCH₂I with aryl-substituted alkenes.

Entry	Phenol	Equiv	Conversion of alkenes [%] ^[a]		
			14 ^[b]	15 ^[c]	16 ^[d]
1	1	2.0	22	10	< 1
2	2	2.0	45	20	24
3	3	2.0	60	26	35
4	4	2.0	–	–	44
5	5	2.0	> 95	> 95	> 95
6	6	2.0	> 95 (94)	> 95 (91)	> 95 (94)
7	7	2.0	> 95	95	88
8	8	2.0	57	30	45
9	9	2.0	87	27	20
10	10	2.0	66	18	15
11	11	2.0	> 95	85	91
12	11	1.1	89	73	95
13	12	2.0	> 95	95	94
14	12	1.1	> 95	79	94
15	13	2.0	> 95	89	> 95
16	EtZnCH ₂ I	2.0	85	50	58
17	Zn(CH ₂ I) ₂	1.0	86	47	50
18	Zn(CH ₂ Cl) ₂	1.0	> 95	> 95	93

[a] Yields of isolated, analytically pure cyclopropanes are in parentheses.

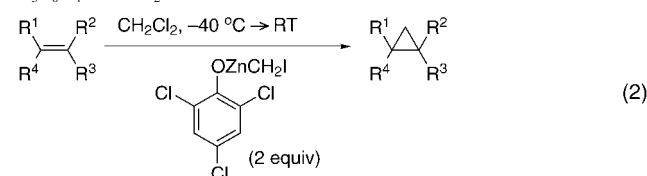
[b] **14**: R¹ = CH₃, R² = R³ = H. [c] **15**: R¹ = R² = R³ = H. [d] **16**: R¹ = R² = H, R³ = CH₂-Ar.

The reactivity of the reagents is highly dependant upon the position and nature of the substituents on the aromatic ring. The first observation that can be made is the apparent need for two *ortho* substituents to achieve high conversion into the corresponding cyclopropyl derivative (entries 1–3 versus entries 5, 7, 11). This trend is a consequence of the self-destruction of the reagent by an intramolecular electrophilic aromatic substitution,^[13] which is otherwise prevented by *ortho* substitution. Another striking feature of these reagents is the beneficial effect of having a strong electron-withdrawing group at the para position (entries 9–11). Although a clear trend between the pK_a of the phenol and the yield of cyclopropanation has not been established, it is obvious that having several electron-withdrawing groups is suitable. For example, 2,4,6-trihalophenol-derived reagents **5**–**7** (when X = F, Cl, and Br, respectively) produced very high yields of cyclopropanes. This higher reactivity could result from an increase in the electrophilicity of the zinc carbenoid and a decrease in the nucleophilicity of the aromatic ring (thus

preventing the rapid decomposition of the carbenoid). Of special interest are the high reactivities of the carbenoids prepared from phenols **11** and **12**. Excellent conversions are observed even when only 1.1 equivalents of the carbenoid are used (entry 11 versus 12, and 13 versus 14). It is important to note that the phenol by-product can be easily recovered in nearly quantitative yield by flash chromatography or by acid-base extraction after the reaction. The reagent derived from 2,4,6-trichlorophenol was more extensively studied since it displayed a very high reactivity for the cyclopropanation reaction and the precursor is cheap and readily available. Direct comparison between the reactivity of traditional zinc reagents and the 2,4,6-trichlorophenol-derived reagent indicates that the latter is an equal or a better reagent for the cyclopropanation of unfunctionalized olefins when the same number of active methylene group equivalents are used (entry 6 versus 16–18). The zinc phenoxide reagent is typically more reactive than Furukawa et al.'s reagent^[8] and bis(iodomethyl)zinc and of comparable reactivity to Denmark and Edwards' bis(chloromethyl)zinc.^[9]

In addition to aryl-substituted olefins, alkyl-substituted alkenes are also converted into the corresponding cyclopropane in high yields (Table 2). The yields are excellent in all

Table 2. Cyclopropanation of alkyl-substituted alkenes with 2,4,6-Cl₃C₆H₃OZnCH₂I.



Entry	Alkene	Conversion [%] ^[a]
1		> 95 (90)
2		> 95 (97)
3		> 95 (93) ^[b]
4		> 95 (96)
5 ^[c]		> 95 (98)
6 ^[c]		92 (92 ^[d] , 85 ^[e])

[a] Yields of isolated products are given in parentheses. [b] In this case, 3 equiv of the carbenoid were used. [c] Bn = Benzyl. [d] 1 equiv of the carbenoid was used (see text). [e] 4 equiv of the carbenoid was used (see text).

cases. As observed with other zinc reagents, it is also possible to achieve a chemoselective monocyclopropanation of an allylic ether in the presence of a trisubstituted olefin when only one equivalent of the reagent is used (entry 6). The use of an excess of the reagent led to the dicyclopropane derivative. Although we have not been able to characterize the reagent by NMR spectroscopy yet, we have shown that a solution of 2,4,6-Cl₃C₆H₃OZnCH₂I in CH₂Cl₂ is stable for at least one hour at 0 °C, which indicates that the reagent is quite stable. However, a significant drop in the yield was observed when a solution of the reagent that was four hours old was used.

In conclusion, the iodomethylzinc phenoxides offer a valuable alternative to the traditional Simmons–Smith reagents due to their ease of preparation, high reactivities towards unfunctionalized olefins, and good stabilities. Further work is currently underway to study the behavior of these carbenoids in directed reactions and to develop an efficient asymmetric cyclopropanation of unfunctionalized olefins.^[14, 15]

Experimental Section

Typical procedure for the cyclopropanation reaction using iodomethylzinc 2,4,6-trichlorophenoxide: Et₂Zn (103 μL, 1.0 mmol) was added to a solution of 2,4,6-trichlorophenol (0.197 g, 1.0 mmol) in CH₂Cl₂ (10 mL) at –40 °C. The solution was stirred for 15 min at that temperature and then CH₂I₂ (81 μL, 1.0 mmol) was added. After stirring for an additional 15 min, α-methylstyrene (65 μL, 0.5 mmol) was added. The cold bath was removed and the mixture was stirred for 12 h. Pentane (25 mL) was added and the organic phase was washed sequentially with 10 % aq HCl (2 × 25 mL), saturated aq NaHCO₃ (2 × 25 mL), saturated aq Na₂SO₃ (25 mL), and saturated aq NaCl (25 mL). The organic layer was dried over MgSO₄ and concentrated to approximately 300 μL under reduced pressure. Flash chromatography (100 % pentane) afforded the desired cyclopropane (62 mg, 94 %) as a colorless liquid: *R*_f = 0.47 (pentane); ¹H NMR (400 MHz, CDCl₃): δ = 7.39 (m, 4H), 7.28 (m, 1H), 1.54 (s, 3H), 0.99 (dd, *J* = 5.9, 4.4 Hz, 2H), 0.85 (dd, *J* = 6.1, 4.2 Hz, 2H); ¹³C NMR (100 MHz, CDCl₃): δ = 147.1, 128.3, 126.8, 125.5, 25.8, 19.8, 15.7; HR-MS: calcd for C₁₀H₁₂: 132.093900, found: 132.092033; elemental analysis: calcd for C₁₀H₁₂: C 90.85, H 9.15; found: C 90.87, H 9.30.

Received: July 10, 2000

Revised: October 4, 2000 [Z15426]

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A Near-Infrared Luminescent Label Based on Yb^{III} Ions and Its Application in a Fluoroimmunoassay**

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There has been a great deal of interest in luminescent lanthanide complexes since Weissman's discovery that in these complexes "the excitation may be accomplished, under suitable conditions, through light absorption by other constituents of the rare earth compound with subsequent transfer of energy to the rare earth ion".^[1] The energy transfer from organic chromophores to lanthanide ions that he described provides an effective way to excite the long-lived, sharply spiked emission. Direct excitation of lanthanide ions is difficult because of the forbidden nature of the electronic transitions in these ions. One of the most important applications of lanthanide complexes is as luminescent labels in clinical diagnostics where they provide an alternative to radioactive probes.^[2–5] Since lanthanide luminescence decays much more slowly than the autofluorescence from the biological material, the latter is suppressed when using time-gated detection.

Research on luminescent lanthanide complexes has been almost exclusively devoted to europium(III) and terbium(III) complexes.^[6–8] The energetics of sensitized luminescence, however, dictate that those complexes must be excited with ultraviolet light,^[9] which is not desirable when working with vital biosystems, requires special optics, and causes extensive scattering in inhomogeneous media. Although some recent work has demonstrated that the excitation window for Eu^{III} complexes can be extended to the violet edge of the visible region^[10] or even to the blue region,^[11] it would be desirable to use significantly longer wavelengths for excitation. Lanthanide ions that emit in the near-infrared, such as ytterbium(III),^[12] neodymium(III),^[13] and erbium(III), may be excited through organic dyes that absorb at wavelengths extending towards the red region of the spectrum. With their potential use as luminescent labels in mind we designed and studied well-defined, water-soluble near-infrared luminescent lanthanide complexes containing organic dyes as the light-absorbing unit.^[14] Recently, we found that 4',5'-bis[*N,N*-bis(carboxymethyl)aminomethyl]-fluorescein (fluorexon, Fx) is a very efficient sensitizer and a strongly binding ligand for near-infrared-luminescent lanthanide ions.^[15] The thermodynamic stability of the $[\text{Yb}(\text{Fx})]$ complex is comparable to that of $[\text{Yb}(\text{EDTA})]$ (EDTA = ethylenediaminetetraacetate).

Here we report on the ligand FxITC (**5**; Scheme 1), which is structurally similar to Fx but carries an isothiocyanate group (ITC) that is reactive towards amine groups and so can be coupled to proteins. The iminodiacetic acid groups ensure firm complexation of lanthanide ions (like they do in $[\text{Yb}(\text{Fx})]$), and the dichlorofluorescein chromophore acts as a sensitizer to near-infrared lanthanide luminescence.

The synthesis of FxITC (Scheme 1) reflects the idea of combining the syntheses of Fx^[16] and fluorescein isothiocyanate.^[17] 2',7'-Dichloro-4-nitrofluorescein diacetate was obtained by condensing 4-nitrophthalic acid with 4-chlororesorcinol at 240 °C and was separated from the 5-nitro isomer by fractional crystallization from acetic acid.^[18] The dichloro derivative was chosen to prevent formation of isomers during the introduction of the methyleneiminodiacetic acid groups. Moreover, the "heavy" chlorine atoms might enhance intersystem crossing in the chromophore which is favorable for sensitizing lanthanide luminescence.^[15]

Direct introduction of the methyleneiminodiacetic acid groups, as used in the production of Fx,^[16] was not successful. Reaction of iminodiacetic acid dimethyl ester^[19] with **2**^[20] and subsequent hydrolysis of the ester yielded **3**, the desired nitro precursor to FxITC. Reduction of **3** using $\text{H}_2/\text{Raney Ni}$ proceeded cleanly, but gave rise to the dinickel complex of **4**, which can be observed with electrospray mass spectrometry. Therefore, **3** was reduced using $\text{Na}_2\text{S}/\text{NaHS}$ in dilute NaOH . After purification by preparative HPLC (C_8 column, eluent: water:acetonitrile:formic acid (155:45:1)), **4** was converted into FxITC using thiophosgene in acetone.

The isopropylamine adduct **6** shows the same complexation behavior towards lanthanide ions as Fx does.^[15] Fluorimetric titrations with Yb^{III} ions indicate formation of a 1:1 complex as long as there is no excess Yb^{III} ions. Excess lanthanide ions leads to the formation of poorly emitting aggregates that so far have escaped further characterization. Such aggregates,

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[**] This work was supported by Akzo Nobel Central Research (Arnhem, The Netherlands). Dr. Fokke Venema and Dr. Harrie Kreuwel of Organon Teknika B.V. (Boxtel, The Netherlands) are gratefully acknowledged for discussions and suggestions regarding the diagnostic test, and for supplying the immunochemicals.