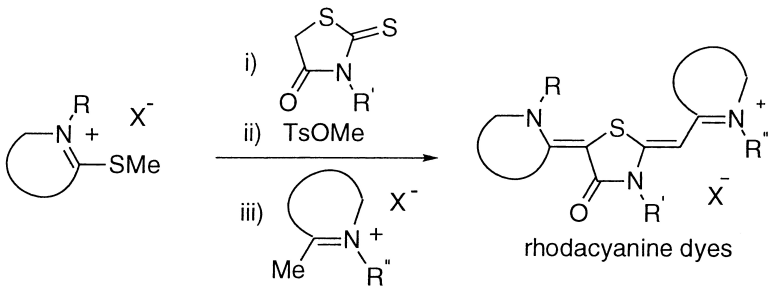
JOURNAL OF combinatorial CHEMISTRY

Report

Parallel Synthesis of Antimalarial Rhodacyanine Dyes by the Combination of Three Components in One Pot

Kiyosei Takasu, Hiroki Terauchi, Hiroshi Inoue, Hye-Sook Kim, Yusuke Wataya, and Masataka Ihara

J. Comb. Chem., **2003**, 5 (3), 211-214• DOI: 10.1021/cc020119z • Publication Date (Web): 04 April 2003



More About This Article

Additional resources and features associated with this article are available within the HTML version:

- Supporting Information
- Links to the 1 articles that cite this article, as of the time of this article download
- Access to high resolution figures
- Links to articles and content related to this article
- Copyright permission to reproduce figures and/or text from this article

View the Full Text HTML

Journal of Combinatorial Chemistry is published by the American Chemical Society.
1155 Sixteenth Street N.W., Washington, DC 20036

Parallel Synthesis of Antimalarial Rhodacyanine Dyes by the Combination of Three Components in One Pot

Kiyosei Takasu,*[,†] Hiroki Terauchi,[†] Hiroshi Inoue,[†] Hye-Sook Kim,[‡] Yusuke Wataya,[‡] and Masataka Ihara*[,†]

Department of Organic Chemistry, Graduate School of Pharmaceutical Sciences, Tohoku University, Aobayama, Sendai 980-8578, Japan, and Faculty of Pharmaceutical Sciences, Okayama University, Tsushima, Okayama 700-8530, Japan

Received December 27, 2002

Malaria is one of the most serious parasitic diseases, in addition to AIDS (HIV) and tuberculosis. According to the recent WHO report, 300−500 million people are infected with the disease and 1.5−2.7 million people, mostly infants or young children, die each year in tropical and subtropical zones. Even the temperate zones will be exposed to the danger of infection, owing to problems such as global warming and traffic globalization. In the last few decades, rapid and widespread movement of drug-resistant malaria parasites has reduced the effectiveness of commonly used chemotherapeutic agents.[2,3] Thus, the development of a novel class of antimalarial compounds has become essential and a worldwide issue. In this regard, new antimalarials, displaying different mechanisms of action in the patients infected with drug-resistant parasites will gain more prominence.[4]

We have recently discovered that rhodacyanine dyes **1** show strong in vitro antimalarial activitiy against *Plasmodium falciparum* (Figure 1).[1] As a result of our preliminary structure−activity relationship (SAR) study, MKH-57 (**1a**), a member of this family, was found to display high antimalarial activity and a significantly good selective toxicity. In light of this finding, we envisioned that a chemical library of cationic rhodacyanine dyes would be useful as a tool to understand both their mechanism of action and SAR. We have planned to develop a combinatorial synthesis of rhodacyanine dyes to provide a diverse compound library for an antimalarial screening test. Since it is already known that the dyes **1** can be used as anticancer agents,[5,6] fluorescent dyes,[7] and photo sensitizers,[8] a new study on the library of rhodacyanine compounds would offer much more useful information. In this Report, we describe a one-pot parallel synthesis of antimalarial rhodacyanines by combining three components in a solution phase and studying their antimalarial activities.[9]

In general, the rhodacyanine dyes **1** have two different conjugated systems, a neutral merocyanine and a cationic cyanine moiety, consisting of three heterocyclic components, in which two end heteroaromatic rings, α and γ, flank a rhodanine moiety β (Figure 1). On the basis of the previously reported strategies,[1,5] these dyes **1** can be obtained with ease using a three-step synthetic sequence: (i) merocyanine formation by condensation of methylthioiminium salt **2** with rhodanine **3** to afford **4**, (ii) activation by S-alkylation of **4** to give the corresponding thioimidate cation **5**, and (iii) cyanine formation by condensation of **5** with methyliminium salt **6** (Scheme 1). All three processes proceed in a very clean fashion that requires only simple operations throughout the reaction and purification steps. Finally, the pure rhodacyanines could be obtained in good yield. Notably, all synthetic intermediates in every stage (i)−(iii) can be purified by simple crystallization and filtration without any chromatographic handling. We thought that the use of a heat-resistant filter tube, a syringe-type vessel equipped with a microfilter at bottom side, would offer much easier operation throughout the synthetic sequence; hence, a one-pot multistep synthesis of rhodacyanines could be accomplished.

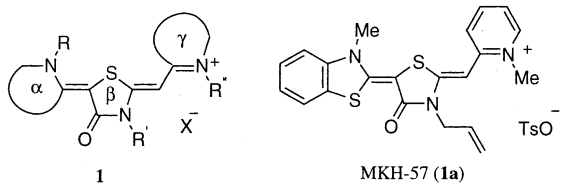
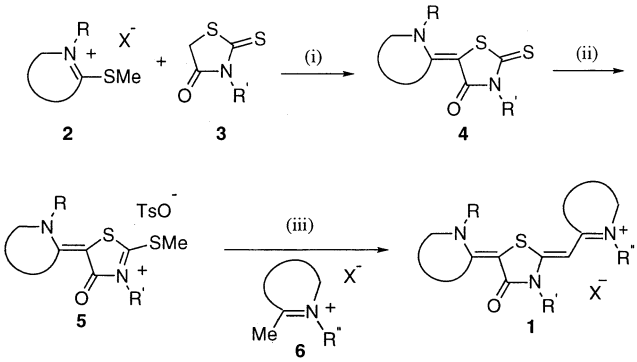


Figure 1. General formula of the rhodacyanine dyes and MKH-57 (**1a**).

Scheme 1. General Synthetic Procedures of **1**[a]



[a] (i) NEt_3, CH_3CN, rt; (ii) TsOMe, DMF, 120 °C; (iii) **6**, NEt_3, CH_3CN, 70 °C.

Table 1. Chemical Yields, EI-MS Results (M^+), and Absorption Properties of the Merocyanine Dye Library[a,b]

	3		
2	**3a**	**3b**	**3c**
2a	**4a**	**4b**	**4c**
	81% (M^+ 246)	84% (M^+ 308)	80% (M^+ 316)
	λ_{abs} 396.5 nm	λ_{abs} 398.0 nm	λ_{abs} 398.0 nm
2b	**4d**	**4e**	**4f**
	92% (M^+ 328)	90% (M^+ 390)	94% (M^+ 398)
	λ_{abs} 430.0 nm	λ_{abs} 432.0 nm	λ_{abs} 431.5 nm
2c	**4g**	**4h**	**4i**
	91% (M^+ 344)	89% (M^+ 406)	96% (M^+ 414)
	λ_{abs} 444.0 nm	λ_{abs} 445.5 nm	λ_{abs} 445.0 nm

[a] Purities of all synthetic merocyanines **4** were 95% or more (estimated by ^1H NMR). [b] UV−vis spectra of the dyes **4** were measured as $CHCl_3$ solution.

† Tohoku University.
‡ Okayama University.

10.1021/cc020119z CCC: $25.00
Published on Web 04/04/2003

Chart 1

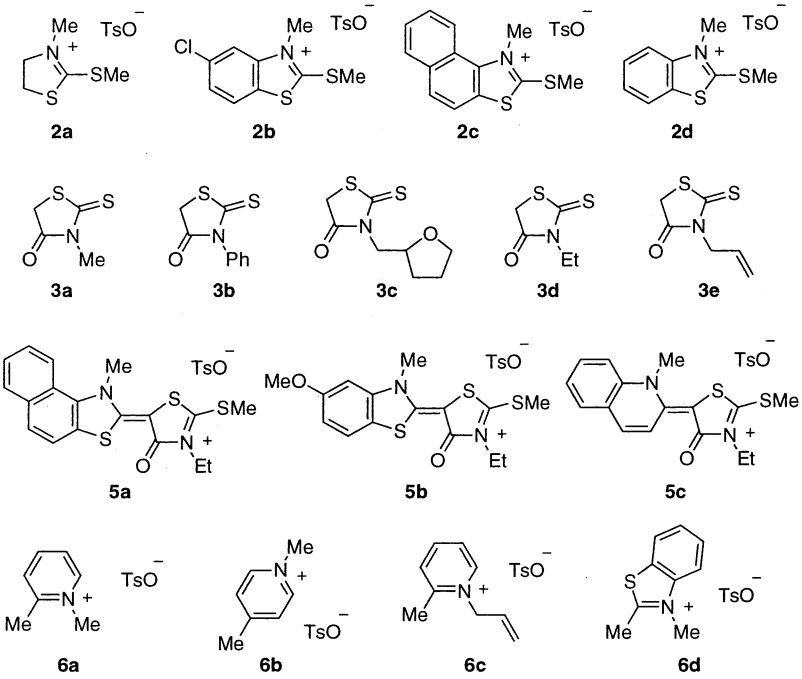
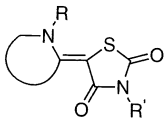


Chart 2



Initially, we optimized each step in this process from (i) to (iii), separately, using the filter tube equipment. As a first step, a combinatorial synthesis of different merocyanines **4** was carried out. To an equimolar mixture of methylthioiminium **2** and rhodanine **3** in a filter tube capped with a septum at the bottom end was added a solution of triethylamine in acetonitlile. After being stirred for 2 h at ambient temperature, the desired merocyanine was obtained as a precipitate. By opening the bottom cap, the CH_3CN solvent was removed through the filter. The precipitate was washed with CH_3CN twice and dried in vacuo overnight to afford merocyanine **4**. Thus, all merocyanine compounds **4a**−**i** were obtained from the combination of **2a**−**c** and **3a**−**c** (Chart 1) in 80∼96% isolated yield and >95% purity (Table 1).[10]

In the next stage of our synthetic plan, we observed that the S-methylation reaction could be successfully carried out in the filter tube apparatus to give **5** in quantitative yield.[10] To a suspension of merocyanine **4** in distilled DMF was added methyl *p*-toluenesulfonate, and the mixture was heated for 4 h at 120 °C. The precipitate was collected and washed with acetone to give **5** as a solid. In this step, it is very important to avoid contamination of water; otherwise, side reactions will occur to furnish oxothiazolines **7** (Chart 2), which are hard to separate from **5**.

Finally, this combinatorial approach was validated by the cyanine formation reaction (Table 2). Nine rhodacyanines **1b**−**j** from a combination of **5a**−**c** and **6a**−**c** were obtained in analytically pure forms (>95% purity) by a procedure similar to that described for the preparation of mericyanines

Table 2. Chemical Yields, FAB-MS Results (M^+), and Absorption Properties of Rhodacyanine Dye Library[a,b]

	6		
5	**6a**	**6b**	**6c**
5a	**1b**	**1c**	**1d**
	41% (M^+ 432)	31% (M^+ 432)	39% (M^+ 458)
	λ_{abs} 505.0 nm	λ_{abs} 519.5 nm	λ_{abs} 514.5 nm
5b	**1e**	**1f**	**1g**
	49% (M^+ 412)	57% (M^+ 412)	39% (M^+ 438)
	λ_{abs} 500.5 nm	λ_{abs} 514.5 nm	λ_{abs} 507.5 nm
5c	**1h**	**1i**	**1j**
	69% (M^+ 376)	59% (M^+ 376)	74% (M^+ 402)
	λ_{abs} 546.0 nm	λ_{abs} 560.5 nm	λ_{abs} 543.5 nm

[a] Purities of all synthetic rhodacyanines **1** were 95% or more (estimated by ^1H NMR). [b] UV−vis spectra of the dyes **1** were measured as MeOH solution.

4. The isolated yields in this combinatorial synthesis were medium, ranging from 31 to 74% yield; however, the yields compare favorably with the yield in the synthesis of MKH-57. It is interesting to note that the merocyanines **4a**−**i** and rhodacyanines **1b**−**j** display a range of colors in the solid state as well as in the solution phase, depending on their partial structure. The absorption maximums (λ_{max} = 397−561 nm) of merocyanines in CH_2Cl_2 and rhodacyanines in MeOH are shown in Tables 1 and 2, respectively.

Having established the reaction conditions for each step, we next examined a single-pot reaction for all three steps. Following the optimized conditions, we conducted the 3 × 3 × 3 parallel synthesis using methylthioiminiums **2b**,**c**,**d**, rhodanines **3c**,**d**,**e**, and iminiums **6a**,**c**,**d** as substrates to furnish 27 different kinds of rhodacyanines **1** (Table 3). All rhodacyanines except for **1dca** and **1dcc** were obtained in moderate to good overall yields, in comparison with the reported overall yields (∼40%).[1] The poor yielding of **1dca** and **1dcc** might be caused by their poor ability to crystallize. Their purities were acceptable in the combinatorial chemistry. A characteristic feature of our method, as compared to

Table 3. Summary of One-pot 3 Components Combinatorial Synthesis; Chemical Yields and Purities (parentheses)[a]

	3			
2	**3c**	**3d**	**3e**	**6**
2b	**1bca**	**1bda**	**1bea**	**6a**
	22% (>95%)	28% (>95%)	27% (>95%)	
	1bcc	**1bdc**	**1bec**	**6c**
	36% (>95%)	18% (>95%)	26% (>95%)	
	1bcd	**1bdd**	**1bed**	**6d**
	47% (>95%)	44% (>95%)	49% (85%)	
2c	**1cca**	**1cda**	**1cea**	**6a**
	33% (>95%)	54% (80%)	33% (>95%)	
	1 ccc	**1cdc**	**1cec**	**6c**
	26% (>95%)	53% (80%)	25% (90%)	
	1 ccd	**1cdd**	**1ced**	**6d**
	56% (>95%)	64% (>95%)	53% (>95%)	
2d	**1dca**	**1dda**	**1a** (MKH-57)	**6a**
	3% (90%)	34% (>95%)	26% (>95%)	
	1dcc	**1ddc**	**1dec**	**6c**
	3% (80%)	30% (>95%)	26% (>95%)	
	1dcd	**1ddd**	**1ded**	**6d**
	63% (>95%)	58% (>95%)	60% (90%)	

[a] Purities were estimated by ^{1}H NMR.

conventional combinatorial technology, is that neither polymer-supported material nor chromatographic purification is necessary. Using this combinatorial technique, a number of rhodacyanines could be prepared in large quantity.

The antimalarial activities of the selected synthetic compounds were evaluated in vitro against *P. falciparum* (chloroquine-sensitive FCR-3 strain), and their cytotoxicities were determined against mouse mammary tumor FM3A cells.[11] Selective toxicities, defined by the ratio EC_{50}(FM3A)/EC_{50}(*P. falciparum*), were determined (Table 4). Several rhodacyanines exhibit in vitro antimalarial activity as strong as traditional antimalarial drugs, such as chloroquine. In accordance with the earlier structure−activity relationship (SAR) study, the introduction of too many hydrophobic aromatic rings into the rhodacyanine skeleton leads to a decrease in selective toxicity.[1] For example, although compound **1d** has the most potent antimalarial activities, it shows fairly high toxicity. Finally, we conclude that, among the several rhodacyanines we prepared, compounds **1e** and **1f**, which have a methoxy substituent on the α unit, show a very good antimalarial activity that is comparable to our previously reported compound **1a** (MKH-57).

In summary, we have developed a one-pot three-step combinatorial synthesis of rhodacyanines **1** by the sequential condensation of three components: methylthioiminiums **2**, rhodanines **3**, and iminiums **6**. It is noteworthy that the method requires neither solid support nor chromatographic purification. Among the synthesized rhodacyanines, **1e** and **1f** exhibit good antimalarial efficacy. We believe this work will provide a wide and rapid survey of the biological activities and physical properties of rhodacyanine dyes.

Acknowledgment. We gratefully thank Drs. Tadao Shishido and Makoto Suzuki (Fuji Photo Film Company) for their discussions about the syntheses of rhodacyanines. We also thank Prof. Yoshinori Kondo (Tohoku University) for his suggestions about the combinatorial techniques. This work was supported in part by a Grant-in-Aid for Exploratory Research (no. 13877378) from the Ministry of Education, Culture, Sports, Science and Technology, Japan.

Table 4. Antimalarial Efficacies in Vitro of Selected Synthetic Rhodacyanines **1**

	EC_{50} (M)		
compd	P. falciparum	FM3A	selectivity
1b	1.8×10^{-8}	6.4×10^{-7}	36
1c	5.4×10^{-8}	5.6×10^{-7}	10
1d	6.0×10^{-9}	5.1×10^{-7}	85
1e	1.4×10^{-8}	1.2×10^{-5}	860
1f	2.3×10^{-8}	$>1.5 \times 10^{-5}$	>650
1g	1.1×10^{-8}	3.6×10^{-9}	0.3
1h	2.4×10^{-7}	2.8×10^{-5}	120
1i	1.3×10^{-7}	$>1.7 \times 10^{-5}$	>130
1j	1.0×10^{-8}	2.1×10^{-6}	210
1dda[a]	2.3×10^{-8}	1.1×10^{-5}	480
1ddd[a]	2.6×10^{-7}	7.2×10^{-7}	3.0
1a (MKH-57)[a]	1.2×10^{-8}	1.2×10^{-5}	1,000
1dec	5.2×10^{-8}	5.8×10^{-7}	11
chloroquine	1.8×10^{-8}	3.2×10^{-5}	1,800

[a] See ref 1.

Supporting Information Available. General experimental procedures and characterization data of selected compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) This work is Part 2 in the series "Antimalarial rhodacyanines". Preliminary communication. Takasu, K.; Inoue, H.; Kim. H.-S.; Suzuki, M.; Shishido, T.; Wataya, Y.; Ihara, M. *J. Med. Chem.* **2002**, *45*, 995−998.

(2) Rosenthal, P. J. *Antimalarial Chemotherapy*; Humana Press: Totowa, 2001.

(3) Peters, W. *Brit. Med. Bull.* **1982**, *38*, 187−192. (b) Wernsdorfer, W. H.; Payne, D. *Pharmacol. Ther.* **1991**, *50*, 95−121. (c) White, N. *J. Antimicrob. Chemother.* **1992**, *30*, 571−585.

(4) Olliaro, P. L.; Yuthavong, Y. *Pharmacol. Ther.* **1999**, *81*, 91−110. (b) Ridley, R. G. *Nature* **2002**, *415*, 686−693.

(5) Kawakami, M.; Koya, K.; Ukai, T.; Tatsuta, N.; Ikegawa, A.; Ogawa, K.; Shishido, T.; Chen, L. B. *J. Med. Chem.* **1997**, *40*, 3151−3160. (b) Kawakami, M.; Koya, K.; Ukai, T.; Tatsuta, N.; Ikegawa, A.; Ogawa, K.; Shishido, T.; Chen. L. B. *J. Med. Chem.* **1998**, *41*, 130−142.

(6) MKT-077, one of rhodacyanine dyes, has been subjected to phase I clinical investigation for the treatment of solid tumors. (a) Propper, D. J.; Braybrooke, J. P.; Taylor, D. J.; Lodi, R.; Styles, P.; Cramer, J. A.; Collins, W. C. J.; Levitt, N. C.; Talbot, D. C.; Ganesan, T. S.; Harris, A. L. *Ann. Oncol.* **1999**, *10*, 923−927. (b) Britten, C. D.; Rowinsky, E. K.; Baker, S. D.; Weiss, G. R.; Smith, L.; Stephenson, J.; Rothenberg, M.; Smetzer, L.; Cramer, J.; Collins, W.; Von Hoff, D. D.; Eckhardt, S. A. *Clin. Cancer Res.* **2000**, *6*, 42−49.

(7) Jeffreys, R. A.; Knott, E. B. *J. Chem. Soc.* **1952**, 4632−4637. (b) Knott, E. B.; Jeffreys, R. A. *J. Chem. Soc.* **1952**, 4762−4775.

(8) van Dormael, A. E. *Ind. Chim. Belge* **1953**, *18*, 1297−1302.

(9) Only limited applications to the combinatorial synthesis in the dye chemistry have been reported. For examples, see: (a) merocyanines: Würthner, F.; Sens, R.; Etzbach, K.-H.; Seybold, G. *Angew. Chem. Int. Ed.* **1999**, *38*, 1649−1652. (b) cyanines: Iacsson, J.; Westman, G. *Tetrahedron Lett.* **2001**, *42*, 3207−3210. (c) courmarine-type OLED: Schiedel, M.-C.; Briehn, C. A.; Bäuerle, P. *Angew. Chem. Int. Ed.* **2001**, *40*, 4677−4680. (d) azo dyes: Merrington, J. M.;

James, M.; Bradley, M. *Chem. Commun.* **2002**, 140−141. (e) conjugated polymer-type OLED: Lavastre, O.; Illitchev, I.; Jegou, G.; Dixneuf, P. H. *J. Am. Chem. Soc.* **2002**, *124*, 5278−5279.

(10) Although there are several possible geometrical isomers of rhodacyanines **1** and merocyanines **4**, the structures shown in the Schemes are depicted as a single geometrical isomer. It is well-known that the conjugated double bonds can be easily isomerized in the solution, and the trans geometrical isomers are usually thermodynamic products.

(11) Kim, H.-S.; Shibata, Y.; Wataya, Y.; Tsuchiya, K.; Masuyama, A.; Nojima, M. *J. Med. Chem.* **1999**, *42*, 2604−2609.

CC020119Z