HANDBOOK OF FLUORESCENT
DYES AND PROBES
Dedicated To

My Wife

Mrs. Madhuri R. Sabnis
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Preface

Fluorescence has been a fascination for individuals for a long time. This book is intended as a reference guide on fluorescent dyes used in medicine, life science, imaging science, cell biology, labeling technology, clinical science, biological chemistry, dye chemistry, biological staining, color chemistry, environmental science, forensic science, organic chemistry, histochemistry, cytochemistry, medicinal chemistry, adhesives, agriculture, coatings, devices, electronics, petroleum, photography, plastics, polymers, security, textile and toys. There are hundreds of fluorescent dyes reported but this book mainly focuses on those dyes, which are widely used in various industrial and academic research.

There is no book available in the market directly on fluorescent dyes and probes, which provides information (such as CAS Registry Numbers, synthesis, various properties, safety/toxicity data and a wide variety of applications) in one source, even though use of fluorescent dyes is wide spread, growing rapidly and has exploded in the past decade. There was a need to publish a book that provided an immediate incentive for compiling the notes to update the scientific community with the wealth of information on fluorescent dyes and probes. To remedy this situation, we have undertaken an ambitious and monumental task of assembling in one publication all the critical data relevant in the field of fluorescent dyes. The dyestuff literature, particularly on fluorescent dyes, is largely in patents. The book provides systematic and up-to-date library of information on 150+ fluorescent dyes and probes as a reference handbook. The book is compiled as a resource guide for chemist and non-chemist in industry as well as in university.

Apart from supplying specific data, the comprehensive, interdisciplinary and comparative nature of the book will provide the user with an easy overview of the state of the art, pinpointing the gaps in the fluorescent dyes knowledge and providing a basis for further research. In addition, it will enable the researcher to use the book in most facile and user-friendly manner.

Fluorescent dyes and probes are arranged alphabetically by the most commonly used name. Again, the choice of primary name is somewhat arbitrary, but an effort has been made to strike a balance between names that are easily recognizable and names that are chemically informative. The detail information of each fluorescent dye or probe is covered in the following order: CAS registry number, chemical structure, CA index name, other names, Merck index number (Merck Index 15th Edition, 2013), chemical/dye class, molecular formula, molecular weight, physical form, solubility, melting point, boiling point, $pK_a$, absorption ($\lambda_{max}$), emission ($\lambda_{max}$), molar extinction coefficient, quantum yield, synthesis, imaging/labeling applications, biological/medical applications, industrial applications, safety/toxicity and references. Where there are discrepancies between different values, the author used his judgment on selecting the most likely value.

Numerous recent references have been provided on various synthetic methods, imaging/labeling applications, biological/medical applications, industrial applications and safety/toxicity data. Space and format limitations prevent giving all the references for each dye. This is the first ever book which provides safety/toxicity data with reference to acute toxicity, aquatic toxicity, carcinogenicity, cytotoxicity, chronic toxicity, ecotoxicity, genotoxicity, hepatotoxicity, immunotoxicity, microbial toxicity, mutagenicity, nephrotoxicity, neurotoxicity, nucleic acid damage, oral toxicity, phototoxicity, phytotoxicity, skin toxicity, reproductive toxicity, and so on.

Several appendixes have been provided at the end of the book for scientists to conveniently and easily find a dye as per their need. These appendixes include CAS Registry Numbers, Acridines, Anthracenes, Boron co-ordination compounds/dyes, Coumarins, Cyanines/Styryls, Heterocycles, Pyrenes and Xanthenes. Omissions as well as errors of fact and interpretation are inevitable in dealing with so vast a subject as fluorescent dyes. I shall be glad to have my attention drawn to errors and to incorporate suggestions for improvement when a revision becomes possible.

I express my profound respect and appreciation to my Guru/Mentor/Advisor, Prof. D. W. Rangnekar, who brought me to this wonderful world of Color Science in the Department of Dyestuffs Technology, Institute of Chemical Technology (ICT), where I laid the foundation stone for my research career in Dye Chemistry.
It is a pleasure to make grateful acknowledgement to Dr. Alan Fanta, Dr. Doina Ene, and Dr. Jeffrey Talkington for extremely useful discussions, encouragement and inspiration.

Words are inadequate to express my sincere appreciation to my wife Madhuri and daughter Anika. It would not have been possible to write this book without their encouragement and patience. It is a great pleasure to express my gratitude and appreciation to John Wiley & Sons Inc., for giving me an opportunity to write this book.

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Ram W. Sabnis is a Senior Patent Agent in USA. His interests include dyes, pigments, organic chemistry, heterocycles, polymers, synthesis, formulations, coatings and patents. Presently, he focuses on drafting and prosecuting US and international patents. He is a registered patent agent with US Patent and Trademark Office (USPTO). Prior to entering the legal (patents) field, he was a research chemist for Ascadia, General Electric, Brewer Science, U.S. Textiles and Thermo Fisher (Molecular Probes) in USA. He also worked as a Patent Agent at Squire Patton Boggs L.L.P. and Senior Manager at Pfizer Inc. (Wyeth).

Dr. Sabnis was born and raised in Mumbai, India. He received his Ph.D. in Organic Chemistry (Dye Chemistry) from Institute of Chemical Technology (ICT) (formerly UDCT), Mumbai, India. He is a Chartered Colourists, Fellow of Society of Dyers and Colourists (CCol FSDC).

Dr. Sabnis is one of the world’s foremost experts in dyes, inventing world’s first colored bubbles (non-staining) and color changing dye system with many applications. He has immensely contributed to color science and technology for the past 25 years, particularly, dyes for biomedicine, personal care products, health/beauty products, electronics (displays, OLEDs), inks, paints, plastics, textiles and toys/bubbles.

He has over 200 publications which include books, book chapters, encyclopedia chapters, patents, reviews, papers, and symposia presentations. He is also an inventor of several US and international patents (issued/published). Dr. Sabnis is the recipient of Pfish Award, Perkin Innovation Award, Grand Innovation Award, Competitive Spirit Award and Best Doctoral Thesis Award. Dr. Sabnis is awarded the Gold Medal for “Outstanding service to the coloration industry” by the Society of Dyers and Colourists, Bradford, U.K.

He has written two books on color chemistry, namely, “Handbook of Biological Dyes and Stains” and “Handbook of Acid-Base Indicators.” He will continue to focus his activities on fascinating dye chemistry as well as demanding intellectual property in the years to come.
ACRIDINE HOMODIMER

CAS Registry Number 57576-49-5

Chemical Structure

\[
\begin{align*}
\text{Cl} & \quad \text{N} \\
& \quad \text{OCH}_3 \\
& \quad \text{Cl} \\
& \quad \text{NH} \\
& \quad \text{(CH}_2\text{)}_3 \\
& \quad \text{NH} \\
& \quad \text{(CH}_2\text{)}_4 \\
& \quad \text{NH} \\
& \quad \text{(CH}_2\text{)}_3 \\
& \quad \text{NH} \\
& \quad \text{N} \\
\end{align*}
\]

CA Index Name 1,4-Butanediamine, \(N,N'\)-bis[3-[(6-chloro-2-methoxy-9-acridinyl)amino]propyl]-

Other Names Acridine homodimer; NSC 219743
Merck Index Number Not listed
Chemical/Dye Class Acridine
Molecular Formula \(C_{38}H_{42}Cl_2N_6O_2\)
Molecular Weight 685.69
Physical Form Orange-brown powder or yellow solid
Solubility Soluble in water, \(N,N'\)-dimethylformamide, dimethyl sulfoxide, methanol
Melting Point 169–170 °C
Boiling Point (Calcd.) 885.4 ± 65.0 °C Pressure: 760 Torr
pKa (Calcd.) 10.63 ± 0.19 Most Basic Temperature: 25 °C
Absorption (\(\lambda_{\text{max}}\)) 431 nm (H\(_2\)O/DNA); 418 nm (MeOH)
Emission (\(\lambda_{\text{max}}\)) 498 nm (H\(_2\)O/DNA); 500 nm (MeOH)
Molar Extinction Coefficient 12,000 cm\(^{-1}\) M\(^{-1}\) (MeOH)
Synthesis Synthetic methods\(^1\)–\(^3\)
Imaging/Labeling Applications Nucleic acids;\(^1\)–\(^9\) chromosomes\(^10\)
Biological/Medical Applications Detecting nucleic acids;\(^1\)–\(^9\) diagnosis and selective tissue necrosis;\(^11\) treating cancer,\(^11\) malformed proteins causing neurodegenerative disease,\(^13\) prion disease\(^12\)
Industrial Applications Not reported
Safety/Toxicity Neurotoxicity\(^13\)

REFERENCES


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ACRIDINE ORANGE (AO)

CAS Registry Number 65-61-2

Chemical Structure

![Chemical Structure of Acridine Orange](image)

CA Index Name 3,6-Acridinediamine, N\(^3\),N\(^3\),N\(^6\), N\(^6\)-tetramethyl-, hydrochloride (1:1)

Other Names 3,6-Acridinediamine, N\(^3\),N\(^3\),N\(^3\),N\(^6\) tetramethyl- monohydrochloride; Acridine Orange R; Acridine, 3,6-bis(dimethylamino)-, hydrochloride; Acridine, 3,6-bis(dimethylamino)-, monohydrochloride; 3,6-Bis(dimethylamino)acridine hydrochloride; Acridine Orange; AO; Acridine Orange N; Acridine Orange NO; Acridine Orange NS; Basic Orange 14; Basic Orange 3RN; C.I. 46005; C.I. Basic Orange 14; Rhoduline Orange NO; Sumitomo Acridine Orange NO; Sumitomo AcridineOrange RK conc

Merek Index Number Not listed

Chemical/Dye Class Acridine

Molecular Formula C\(_{17}\)H\(_{20}\)ClN\(_3\)

Molecular Weight 301.82

Physical Form Orange solid

Solubility Soluble in water, dimethyl sulfoxide, ethanol, methanol

Absorption (\(\lambda_{\text{max}}\)) 500 nm (H\(_2\)O/DNA); 460 nm (H\(_2\)O/RNA); 489 nm (MeOH)

Emission (\(\lambda_{\text{max}}\)) 526 nm (H\(_2\)O/DNA); 650 nm (H\(_2\)O/RNA); 520 nm (MeOH)

Molar Extinction Coefficient 53,000 cm\(^{-1}\) M\(^{-1}\) (H\(_2\)O/DNA); 64,000 cm\(^{-1}\) M\(^{-1}\) (MeOH)

Synthesis Synthetic methods\(^1\)–\(^8\)

Imaging/Labeling Applications Bacteria;\(^9\)–\(^{15}\) blood smears;\(^16\)–\(^{18}\) casein;\(^19\) cells/tissues;\(^20\)–\(^{23}\) chromosomes;\(^24\),\(^25\) endospores;\(^26\) lignin;\(^27\),\(^107\) liposomes;\(^28\) lysosomes;\(^29\)–\(^{35}\) microorganisms;\(^36\)–\(^{38}\) nucleic acids;\(^52\)–\(^{74}\) parasites;\(^75\)–\(^{79}\) sperms;\(^80\),\(^81\) tumors;\(^82\),\(^83\) yeast

Biological/Medical Applications Analyzing/counting/measuring microbialorganisms;\(^39\)–\(^{49}\) analyzing/detecting/identifying nucleic acids;\(^72\)–\(^{74}\) counting/detecting cells/tissues;\(^20\)–\(^{23}\) detecting parasites;\(^75\)–\(^{79}\) measuring phagosome-lysosome fusion;\(^31\),\(^33\) for photodynamic therapy;\(^82\),\(^83\) monitoring atmospheric/indoor bioaerosols;\(^89\),\(^90\) apoptosis assay;\(^91\)–\(^{97}\) cytotoxicity assay;\(^98\),\(^99\) genotoxicity assay;\(^99\) as temperature sensor;\(^100\),\(^101\) dental materials for crowns and bridges

Industrial Applications Adhesives;\(^103\) aluminophosphate crystalline materials;\(^104\) detecting clay particles;\(^105\) display device;\(^106\) evaluating fiber surface characteristics;\(^107\) glass matrixes;\(^108\) imaging material;\(^109\) inks;\(^110\),\(^111\) lasers;\(^112\) recording materials;\(^113\)–\(^{115}\) photoresists;\(^116\),\(^117\) textiles;\(^118\) thin films;\(^119\),\(^120\) tracers for hydrology;\(^121\) wiring boards

Safety/Toxicity Carcinogenicity;\(^123\)–\(^{125}\) cytotoxicity;\(^126\),\(^127\) DNA damage;\(^128\) embryotoxicity;\(^129\) genotoxicity;\(^130\)–\(^{134}\) mutagenicity;\(^135\)–\(^{138}\) photodynamic toxicity;\(^139\) phototoxicity

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133. He, L.; Jurs, P. C.; Custer, L. L.; Harvey, J. S.; Roberts, K. J.; Tweats, D. J.; Keenan, P. O.; Walmsley, R. M. An analysis of results from 305 compounds tested with the yeast RAD54-GFP genotoxicity assay (GreenScreen GC) - including relative predictivity of regulatory tests and rodent carcinogenesis and performance with autofluorescent and colored compounds. Mutagenesis 2007, 22, 409–416.


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Acridine Orange (AO)


ACRIDINE ORANGE 10-DODECYL BROMIDE (DODECYL-ACRIDINE ORANGE (DAO))

CAS Registry Number 41387-42-2
Chemical Structure

CA Index Name Acridinium, 3,6-bis(dimethylamino)-10-dodecyl-, bromide (1:1)
Other Names Acridinium, 3,6-bis(dimethylamino)-10-dodecyl-, bromide; 10-Dodecylacridine Orange Bromide; 3,6-Bis(dimethylamino)-10-dodecylacridinium bromide; AO 10 Dodecylbromide; Acridine orange 10-dodecyl bromide; Dodecyl-Acridine Orange; BDA; D 455; DADAB; DAO
Merck Index Number Not listed
Chemical/Dye Class Acridine

Molecular Formula C_{29}H_{44}BrN_{3}
Molecular Weight 514.59
Physical Form Orange solid
Solubility Soluble in dimethyl sulfoxide, ethanol, methanol
Melting Point >250 °C
Absorption (λ_{max}) 495 nm (MeOH)
Emission (λ_{max}) 520 nm (MeOH)
Molar Extinction Coefficient 87,000 cm^{-1} M^{-1} (MeOH)
Synthesis Synthetic methods
Imaging/Labeling Applications Chloride ions; keratin fibers/hairs; Langmuir-Blodgett (LB) monolayers; micelles; mitochondrial membranes; proteins
Biological/Medical Applications Analyzing chloride ions; characterizing drug binding sites on glycoproteins; ophthalmic devices (intraocular lenses (IOL))
Industrial Applications Determining cationic surfactants; electroluminescent devices; Langmuir-Blodgett films; photographic imaging system; photoresists; semiconductor electrodes; silica-surfactant composite films
Safety/Toxicity No data available

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