MISQUOTATION IN SCIENCE: THE CASE OF QUININE SULPHATE FLUORESCENCE. (CITATION INEXACTE EN SCIENCE: LE CAS DE LA FLUORESCENCE DE SULFATE DE QUININE)

J. David Holmes
National Research Council of Canada,
A.S.K. Service, Suite 904,
789 Don Mills Road,
Toronto, Ontario
M3C 1T5

ABSTRACT

Citations to a key paper in the literature of photochemistry (W.H. Melhuish, J. Phys. Chem., 65, 229 (1961)) were examined to see how accurately the citing authors had used the results of the cited paper. Of 183 papers examined 52 failed in some way to correctly report the results of the cited paper. Of these 45 used Melhuish's number for the quantum yield of fluorescence of quinine bisulphate with the wrong acid concentration. The origin of this error is discussed and the attempts to correct it are traced. The distribution of the group of 183 papers by subject, language and year of publication is examined. (Les citations d'un article fondamental dans le domaine de la photochimie (W.H. Melhuish, J. PHYS. CHEM, 65, 229 (1961)) sont étudiées afin de déterminer la fidélité avec laquelle les auteurs qui le citent en ont utilisé les résultats. Des 183 documents examinés qui citaient cet article, 52 en ont mal interprété, d'une façon ou d'une autre, les résultats. De ces 52 documents, 45 ont utilisé la valeur de Melhuish pour le rendement quantique de la fluorescence de sulfate de quinine, avec la mauvaise concentration d'acide sulfurique. La cause de cette erreur est commentée et diverses solutions sont proposées. La distribution des 183 documents, par sujet, langue et date de publication, est également étudiée.)

INTRODUCTION

One of the first things that one is taught when embarking on historical research is never to believe secondary sources, but always to examine the primary source where possible. An apochryphal experiment in communication demonstrates how messages passed orally from person to person rapidly become distorted beyond recognition. The purpose of this study was to examine the transmission of a simple scientific finding through the journal literature over a fifteen-year period.

All citations to a key paper in the literature of photochemistry (Melhuish, W.H. 1961 Quantum efficiencies of fluorescence of organic substances: effect of solvent and concentration of the fluorescent solute. J. Phys. Chem. 65: 229-235.) were traced using the Science Citation Index[(R)] using the indexes from 1961 up to and including the second quarter of 1975. The citing papers were examined to see exactly what was attributed to the cited paper. In all, 202 citations were traced of which 183 were examined, the others being unavailable to the author.

The key finding of Melhuish's paper (indicated hereafter by WHM) was a value of the quantum yield of fluorescence of quinine bisulphate which was proposed as a standard to be used in the relative determination of other fluorescence quantum yields. A careful series of experiments determined the absolute value of this quantum yield to be 0.51 for a 5×10^{-3}M solution in 1.0 N H_2SO_4 at 25 C and excited at 366nm. (Simply put, when a molecule absorbs ultraviolet light it may lose its excess energy either by re-emitting the light,i.e. fluorescing or by alternate chemical or thermal processes. The fraction of each absorbed photon that is re-emitted as fluorescence is known as the fluorescence quantum yield. This number is an important measure of how significant are the other de-excitation pathways. The quantum yield is measured by exciting the substance to be studied under carefully controlled conditions and measuring the intensity of the light emitted either in an absolute manner, measuring the heat produced by collecting the light in a bolometer, or in a relative way, by comparing the light intensity emitted with that from a standard compound of known quantum yield. Since absolute measurements are exceedingly difficult to perform, most quantum yields are measured relative to a standard compound such as quinine sulphate.) WHM gives detailed descriptions of the method used and reports quantum yields for several other organic fluorescers in various solvent systems. It was shown that quinine bisulphate is a good standard as it is soluble in aqueous media, photostable, insignificantly quenched by oxygen and has a small fluorescence-absorption overlap. A small temperature effect of -0.2 to -0.3% per degree was

---

(R) Science Citation Index is a registered trademark of the Institute for Scientific Information, 325 Chestnut Street, Philadelphia.

noted in the range from 10-40 C.

It is clear from Melhuish's work as well as both earlier and later papers that the value of the quinine quantum yield is dependent upon a number of conditions, notably temperature and concentration (WHM found that the quantum yield rose to 0.55 when the concentration was extrapolated to infinite dilution), as well as possibly solvent system and exciting wavelength. In particular, it has been shown by several workers (Chen 1972, Dawson and Windsor 1968) that the quantum yield is dependent upon the sulphuric acid concentration and that the infinite dilution value drops from 0.55 to 0.51 when going from 1.0N to 0.1N H_2SO_4: a drop of some 6%. The reason for this effect is not entirely clear (Ware and Rothman 1976) and even its presence is disputed by a few authors but it is fair to say that any author wishing to use WHM's value of 0.51 would be advised to use all the conditions of temperature, exciting wavelength, concentration and acid strength used by WHM or run the risk of applying the wrong number. For example, someone using the value of 0.51 at 20 C and in 0.1N H_2SO_4 could be using a value that is 8% too great.

It is not the intention of this paper to debate the relative merits of various values for the quantum yield of fluorescence of quinine sulphate, nor to propose alternate standards, nor advocate different experimental techniques. There are many such papers in the literature, most of which can be found by tracing citations to WHM. Some key papers on the subject are those of Dawson and Windsor (1968), Weber and Teale (1957), Demas and Crosby (1971), Eastman (1969), Fletcher (1969), Heller et al. (1974) as well as the work of Chen and Melhuish. Suffice it to say that the literature records values for the quantum yield of quinine varying from 0.46 (Rusakowicz and Testa 1968) to 0.70 (Scott 1970), but that by far the most widely quoted value is that of Melhuish determined in 1961 and supported by several later independent measurements. Much published work in the literature of photophysics and photobiology depends upon a relative determination of a fluorescence quantum yield and quinine sulphate is probably the most common standard. The importance of such numbers is emphasized by the recent open dispute between two of the leading authorities in the field over the value of the fluorescence quantum yield of diphenylanthracene. (Berlman 1973)

Distribution of Papers Citing Melhuish

The most striking thing about this paper is the number of times that it has been cited since 1961. In the period 1962-1974 there were an average of 14.8 papers citing it per year. This compares with an average of 1.7 citations per authored item per year in the Science Citation Index as a whole. However, the 166 citations received from 1961-1972 still place it a long way down in the list of most cited papers.

According to statistics published by the I.S.I. the most cited paper during this period received 29,655 citations and even the 100th paper on the list received 865 citations (Garfield 1974). Another unusual feature of the distribution of citations to WHM is the lack of any aging effect. In fact, with the exception of 1973, the number of citations shows an upward trend over time, reflecting the general growth in the literature covered by the S.C.I.

The distribution of citing papers by journal of publication follows the usual Bradford-Zipf distribution as shown by a semi-logarithmic plot of journal rank against cumulative number of citations (Brookes 1969). The slope and intercept of such a plot suggests that the total number of citations should be 165. This low figure probably reflects the inhomogeneity of the subject field represented by the citing papers. The most heavily citing journals are the Journal of Chemical Physics, the Journal of the American Chemical Society and the Journal of Physical Chemistry, in that order. Of the 183 papers traced and examined all but 5 (3 French and 2 German) were in English. Of the remaining papers that were not found, 5 were definitely non-English (2 Russian, 1 French, 1 German and 1 Japanese). Ten out of 202 is a surprisingly low proportion of non-English language articles in the physical sciences, where the usual proportion is normally estimated at nearer 40%.

Figure 1

Distribution of citing papers traced according to address of author

Year	U.S.	U.K.	Germany	France	Japan	Israel	Others	Totals
1961	1							1
1962	1	2						3
1963	5	7					1	13
1964	5							5
1965	6	1						7
1966	13	4			1		1	19
1967	10	1					2	13
1968	7	5		1	1		1	15
1969	13	2					1	16
1970	12		1	1	1			15
1971	7	1	2		4	2	2	18
1972	8	3	3		3	3	1	21
1973	6	1	1					8
1974	13	1	2	1	2		3	22
1975	1	1	1	1	2		1	7
Totals	108	29	10	4	14	5	13	183

An inspection of the papers that were traced revealed that of 183 papers 38, or 20.8% originated in non-English-speaking countries (Israel and India were counted as English speaking as far as science is concerned). Therefore 33 papers were published in English despite the fact that English was probably not the first language of the author. This indicates how dominant an influence is the U.S. in this area of science; not only does the U.S. produce 59.0% of these papers but another 18.1% are published in English by non-English speakers. It comes as no surprise to find that of the 4 papers originating in France, 3 of them are published in French. It is interesting to note the gradual diffusion of citations through the scientific world. In the first 4 years only 1 paper out of 22 originated outside the U.S. or U.K., but in the last 4 years 24 out of 58 papers originated outside the U.S. or U.K. until in the first half of 1975 only 29% of the citations came from British or American laboratories. Surprisingly only 2 of the citing articles came from the U.S.S.R. This is a small proportion even allowing for the fact that the S.C.I. may be weak in its coverage of the Soviet literature and that 2 Russian articles were not available and so do not appear in the table.

Figure 2

Articles citing Melhuish classified by subject scope of periodical in which they appeared. Journals classified by the Library of Congress Classification.

Year of Publication	General Science(Q)	Pure Chemistry(QD)	Applied Chemistry(TP)	Biochemistry (QP)
1961		1		
1962		1	2	
1963	4	8	1	
1964		3		
1965	1	4		
1966	3	13	1	1
1967		8		4
1968	2	8	2	1
1969		12		5
1970		13	2	1
1971		11		4
1972		13	2	6
1973		3	1	2
1974		12	5	4
1975*		3		4
Totals	10	113	16	32

*First half of 1975 only. Citation Index not available for second half of 1975.

Figure 2 (continued)

Year of Publication	Pharmaceutical Science(RM,RS)	Physics (QC)	Other	Totals
1961				1 (1)#
1962				3 (3)
1963				13 (13)
1964		2		5 (5)
1965		2		7 (7)
1966		3		21 (19)
1967		1		13 (13)
1968		4		17 (15)
1969				17 (16)
1970	2		1	19 (15)
1971	2	2		19 (18)
1972		1		22 (21)
1973		3		9 (8)
1974	3	2	1	27 (22)
1975*		1	1	9 (7)
Totals	7	21	3	202 (183)

*First half of 1975 only.

#Numbers in brackets refer to articles traced and inspected.

Figure 2 shows the subject spread of the articles citing WHM as classified according to the Library of Congress Classification normally given to the journal containing the article. Although this way of defining the subject of an article is rather arbitrary some interesting trends can be seen. The original article was published in a core physical chemistry journal in a subject area on the fringe of physics. Of WHM's references 14 out of 16 were to chemical or general science journals. Citations to WHM are rapidly picked up in the applied chemistry literature as fluorescence spectroscopy has for some time been a central technique in analytical chemistry. Three years after the publication of the original, citations appear in the physics literature (mostly optics) and continue to come out over the next 10 years as the boundary between photochemistry and photophysics becomes increasingly blurred. Between 1963 and 1968 a number of citing articles appear in interdisciplinary journals such as Nature and Science and in 1966 the first of an increasing number of citations appears in the biochemistry literature, as quantitative fluorescence measurements start to be used in biological systems. It is tempting to suggest that the link between the physical and life science literature is created by the interdisciplinary journals read by both groups of scientists as has been suggested by other citation studies (Narin 1972). However, although the first biochemical citation is one of the first papers to use quantitative fluorescence techniques for conformational studies of proteins, there is no hard evidence of a link to Melhuish via one of the earlier citations in Science or Nature. This is not to say that such a link was not there but merely that no reference was made to it. Certainly the time-lag of 2-3 years is consistent with the adoption of a new technique and the publication delay ensuing. A similar time-lag occurs before the first of several citations from the pharmaceutical literature appears in 1970. Later other citations appear in the medical and chemical engineering literature. By 1974 less than half the citations are found in the original field of physical chemistry.

REASONS FOR CITATION

Figure 3

Reasons for quoting Melhuish's paper (J. Phys. Chem., 65, 229 (1961)).

Reason	Number of papers citing
Reference to quinine sulphate as a quantum yield standard	109
Reference to quantum yield of compound other than quinine	49
Reference to Melhuish's experimental method or application of correction factors	23
Discussion of problems in obtaining fluorescence quantum yields	17
Independent study of quinine quantum yields	6
Proposal for a new quantum yield standard	5
Other*	7

*Includes 2 papers citing for no apparent reason

Figure 3 attempts to analyse the reasons for citing Melhuish's paper. These reasons can be broken down into 6 major categories of which by far the most important is reference to the use of quinine sulphate as a quantum yield standard with 109 citing articles. The next most important reason is reference to the quantum yield of some other compound listed by Melhuish, as determined relative to quinine sulphate in various solvent systems. The importance attached to such quantum yield values and the the controversy surrounding the published numbers is emphasised by the fact that 6 of the citing papers refer to independent determinations of the quinine sulphate value. Of all the citing papers, however, only 2 authors directly dispute Melhuish's value for the quantum yield of quinine. Five of the citing papers propose new substances as standards. However, 4 of them calibrate these new standards against quinine and all of them misquote the solvent conditions used by Melhuish. One final observation is that 2 authors cite Melhuish for no reason at all, so far as the author could determine!

ACCURACY OF QUOTATION

Each citing article was examined to see if the citing author had quoted what he had attributed to Melhuish correctly. Where the citation was to the use of quinine sulphate as a standard, particular attention was paid to the conditions used, especially the acid concentration. Many articles failed to describe these conditions, dismissing them in phrases such as: "...The absolute quantum yields ... were computed from fluorescence spectra ... using quinine sulphate as standard.", but in other cases detailed conditions were given enabling a comparison to be made with those of the article cited.

The first trivial observation is that 3 citing papers managed to mis-spell Melhuish, coming up with "Melhuis", "Melhuisch" and "Melhish" respectively. Figure 4 shows how a critical examination of the citing papers reveals more substantive misquotations. As can be seen, of the 109 citations to the use of quinine as a reference compound, 23 did not specify the conditions used. Of the 86 articles that did give sufficient information, 49, or more than half used the wrong conditions; 45 of them using the wrong acid concentration. In addition there were 3 other misquotations concerning other aspects of the paper. One of these was clearly a misreading of a table of figures, another was an unjustified switching of a solvent, and the last was a citation to a statement that was not made in the original.

It seems that these figures reveal two sorts of misquotation that might be termed random and systematic. The random misquotations might be expected to occur in any such group of citations. As far as can be determined there are 7 of these out of 183 citations (3.8%); 4 concerning quinine and 3 others. The systematic misquotation is involved in the 45 cases that use WHM's standard value for the quinine quantum yield but with another sulphuric acid concentration. By far the greatest number of these 45 papers use 0.1N

Figure 4

Citations to Melhuish analysed by accuracy of citation.

Year of Publication	I	II	III	IV
1961	-	-	-	-
1962	-	-	-	-
1963	4	1	-	-
1964	3	-	-	-
1965	4	2	1	-
1966	10	4	3	3
1967	11	3	5	4
1968	12	-	8	7
1969	10	2	2	2
1970	11	4	6	6
1971	12	3	6	6
1972	13	1	7	7
1973	4	-	4	4
1974	11	1	5	4
1975	4	2	2	2
Totals	109	23	49	45

I. Citations to Melhuish's figure for quinine quantum yield.
II. Papers in I which did not specify conditions.
III. Papers in I which used Melhuish's value incorrectly.
IV. Papers in III which used the wrong acid concentration.

ACCURACY OF QUOTATION (continued)

as opposed to the I.ON H_2SO_4 used by WHM. Whilst such a difference would not significantly affect the results of most of the papers making this mistake, it could alter the conclusions of some; but more importantly, such a high proportion of inaccuracy suggests a widespread failure to read the original paper with care, if at all.

TRACING THE ORIGIN OF THE MISQUOTATIONS

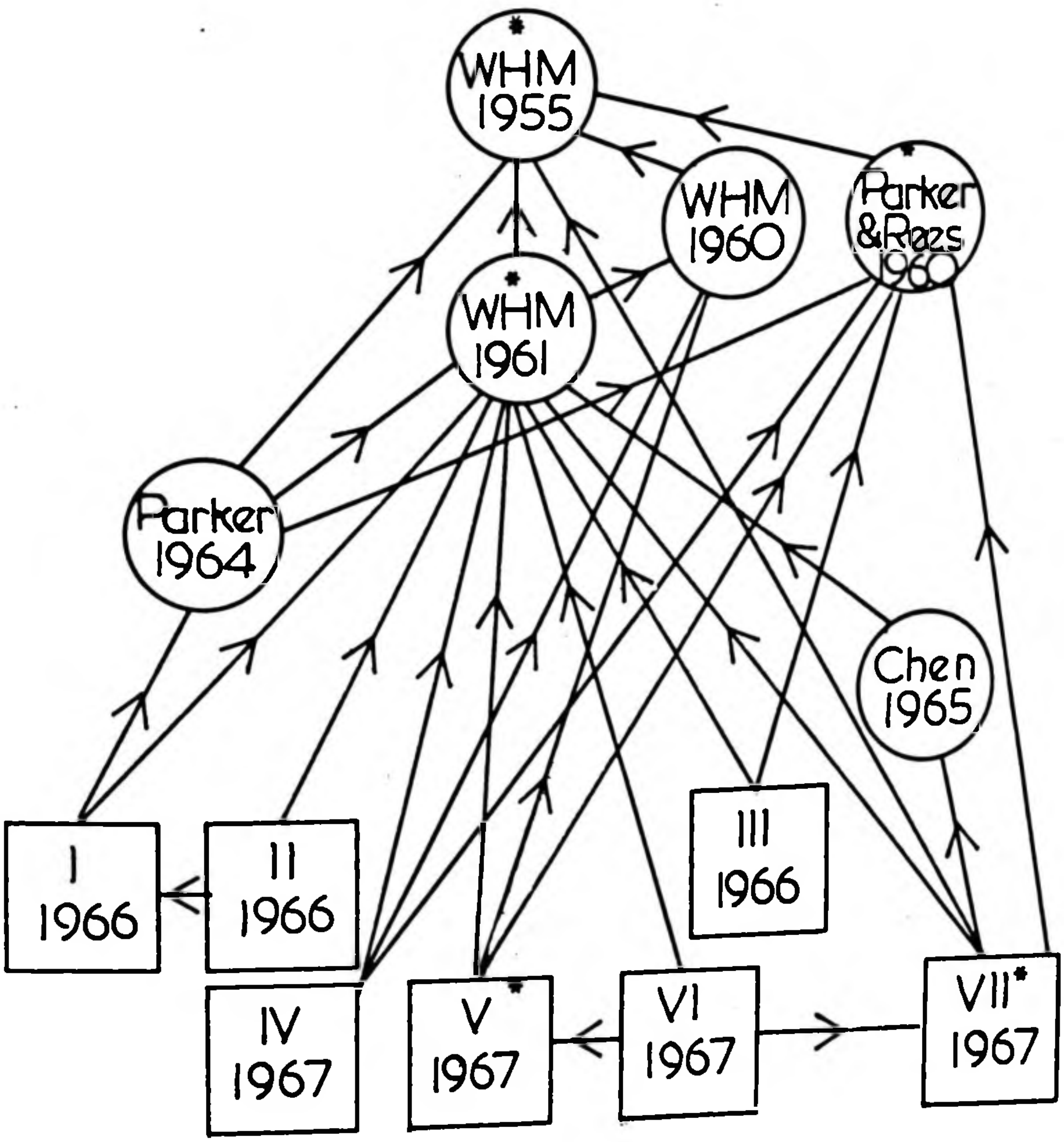
As stated above there seems to be a systematic error in the citations to Melhuish's paper involving the acid concentration used. The first papers to attribute the wrong acid concentration to Melhuish were published in 1966. These 3 papers were examined to see if there was any indication of how this error started. Two of these papers were written by the same principal author and one cites the other regarding the luminescence measurements. Apart from

Melhuish, this author cites a review by C.A. Parker in 1964. In turn this review cites an earlier review by C.A. Parker and W.T. Rees (Analyst, 85, 587 (1960)) which deals with methods of measuring fluorescence quantum yields. In this review they recommend quinine bisulphate in 0.1 N H_2SO_4 as a standard with a quantum yield of 0.55 citing an earlier paper by Melhuish published in 1955 (New Zealand Journal of Science and Technology, 37, 142 (1955)). The third paper using the wrong acid concentration to be published in 1966, which incidentally was also the first paper to be published in a biochemical journal that cites Melhuish, also cites this review by Parker and Rees. The relationships between the 3 papers in 1966 and the 4 in 1967 using the wrong acid concentration, and the earlier papers of Melhuish and Parker are shown diagrammatically in Figure 5.

Of the 4 papers published in 1967 that make this error, 3 quote Parker and Rees and the 4th cites one of the other three. Two of these papers deal specifically with the fluorescence of quinine sulphate (V and VII) and also cite Melhuish's 1955 paper. These 2 papers then help to perpetuate the error as they are cited in turn by later papers. To make matters worse, these two papers are published one in the chemical literature and the other in the biochemical literature. It seems that the misquotation of Melhuish arose due to confusion in co-citing Parker and Rees together with the later Melhuish papers. The error was perpetuated when 2 later papers discussing quinine made this same mistake. The confusion was probably increased by the leading textbook on photochemistry ("Photochemistry", J. G. Calvert and J. N. Pitts, 1966) which appeared in 1966 and recommended quinine as a fluorescence quantum yield standard in 0.1 N acid citing the 1955 Melhuish paper, but not mentioning the later papers.

It is interesting to follow the attempts to correct this misquotation over the next few years. In a review published in 1968 (Dawson and Windsor 1968) note the effect of acid concentration: "... the value of the quantum yield of fluorescence assigned to quinine in 0.1 N H_2SO_4 appears to be incorrect and results in estimates of fluorescence yield that are 6 to 8% too high." They pointed out that the effect of acid concentration on the quantum yield had been reported 7 years earlier in the literature, albeit in German (Eisenbrand 1961). Again in 1971 another review stated (Demas and Crosby 1971): "In spite of the apparent reliability of quinine sulphate large errors may have crept into the literature. Most authors use 0.1 N sulphuric acid solutions of quinine in their measurements yet employ Melhuish's reported quantum yeild for a solution of quinine in 1.0 N sulphuric acid." In 1972 Chen who had published a number of papers with the wrong acid concentration, including paper VII in Fig. 5, wrote: "Many investigators, including ourselves, were guilty of using 0.546 for the yield of quinine in 0.1 N H_2SO_4 rather than 1.0 N acid. We find that the quantum yield actually is 6% lower in the more dilute acid, so a yield of 0.51 should be used." (Chen 1972)

Figure 5



Squares represent the first seven papers to misuse the acid concentration in 1966 and 1967. Circles represent the important earlier papers with the lines representing citation linkages.
* Indicates a paper dealing primarily with fluorescence quantum yield measurements.

Despite these cautions published in widely cited articles papers continue to be published with the same error. Even after the third of these attempts to correct the error in 1972, 10 papers have been published misusing Melhuish's number. It seems to be easier to start a series of systematic misquotations than to correct them!

REFERENCES

BERLMAN, I. 1973 Fluorescence of azulene and 9, 10 diphenylanthracene. A reply to criticism. Chem. Phys. Lett., 21: 344-345.

BROOKES, B.C. 1969 The complete Bradford-Zipf 'bibliograph'. J. Docum, 25: 58-60.

CHEN, R.F. 1967 The fluorescence of quinine. Anal. Biochem., 19: 374-387.

CHEN, R.F. 1972 Measurements of absolute values in biochemical fluorescence spectroscopy. J. Res. Nat. Bur. Stand., 76A: 593-606.

DAWSON, W.R. and WINDSOR, M.W. 1968 Fluorescence yields of aromatic compounds. J. Phys. Chem., 72: 3251-3260.

DEMAS, J.N. and CROSBY, G.A. 1971 The measurement of photoluminescence quantum yields. A review. J. Phys. Chem., 75: 991-1024.

EASTMAN, J.W. 1969 Dependence of fluorescence on solvent and temperature. Spectrochemica Acta 26A: 1545-1557.

EISENBRAND, J. 1961 Fluorimetry V. Quinine sulphate as a fluorescence standard. Z. Anal. Chem., 179: 170-175.

FLETCHER, A.N. 1969 Quinine sulphate as a fluorescence quantum yield standard. Photochem. Photobiol. 9: 439-444.

GARFIELD, E. 1974 Selecting the all-time citation classics. Current Contents, No. 2, Jan. 9: 5-8; The second fifty papers most cited from 1961-1972. No. 7, Feb. 6: 5-9.

HELLER, C.A. et al. 1974 Fluorescence spectra and quantum yields: uranine, 9, 10 diphenylanthracene and 9, 10 - bis (phenylethynyl) anthracenes. J. Chem. Eng. Data, 19: 214-219.

MELHUISH, W.H. 1972 Absolute spectrofluorimetry. J. Res. Nat. Bur. Stand., 76A: 547-560.

REFERENCES

NARIN, F. et al. 1972 Interrelationships of scientific journals. J. Amer. Soc. Inf. Sci., 23: 323-331.

RUJAKOWICZ, R. and TESTA, A.C. 1968 A comparison of quinine bisulphate and 9, 10 -dephenylanthracene as fluorescence standards. J. Phys. Chem., 72: 793-796.

SCHULMAN, S.G. et al. 1974 Fluorescence of 6 - methoxyquinoline, quinine and quinidine in aqueous media. J. Pharm. Sci., 63: 876-880.

SCOTT, T.G. et al. 1970 Emission properties of NADH. J. Amer. Chem. Soc., 92: 687-695.

WARE, W.R. and ROTHMAN, W.C. 1976 to be published.

WEBER, G. and TEALE, F.W.J. 1957 Determination of the absolute quantum yield of fluorescent solutions. Trans. Faraday Soc., 53: 646-655.