

## Paper chromatography of monomeric and dimeric lignin-type sulfonates

With the increased emphasis on elimination of water pollution, research on spent sulfite liquor utilization as an alternative to disposal is increasing. Lignin sulfonates are the primary constituents in the spent liquor usually amounting to 50–60 % or more of the total dissolved solids<sup>1,2</sup>. Of the substances present in the spent liquor, it is the lignin sulfonates that present the most challenge towards quality utilization. Better utilization is dependent upon a better understanding of the lignin sulfonate material.

Macromolecular lignin sulfonate molecules are extremely difficult substances to use in the determination of structure because of the non-repetitive nature of both the basic unit and the chemical linkages involved. As a result, investigations of structure have involved low-molecular-weight lignin sulfonates and lignin-type sulfonate model compounds<sup>3–10</sup>. One of the difficulties in the isolation and purification of low-molecular-weight lignin sulfonates is in the monitoring of the separation procedure and in determining purity of the fractions. A means of alleviating this difficulty is through the use of a discriminatory paper (PC) or thin-layer chromatographic (TLC) procedure. Such an analytical procedure would also be of value in studying the reactions of lignin model compounds under the sulfonation conditions used in the sulfite pulping process.

### Literature review

TLC was used to a limited extent by MOTHERSHEAD AND GLENNIE<sup>9</sup> in the identification of monomeric lignin sulfonates from red alder (*Alnus rubra*). GLENNIE<sup>10</sup> used TLC extensively in studying some of the reactions of monomeric lignin sulfonates. In the latter reference, two different developing solvent systems were used: 1-butanol–water (22:3) and 1-butanol–1-propanol–water–acetic acid (22:18:10:1). Neither developer is very discriminatory with many of the twenty lignin-type sulfonate reference compounds used having identical or nearly identical  $R_F$  values.

PC was used by PARRISH<sup>3,4</sup> in the detection of syringyl-type monomeric lignin sulfonates from the sulfite pulping of wattle wood (*Acacia mearnsii*). Although PARRISH<sup>4</sup> preferred WALEY'S solvent system<sup>11</sup>, 1-butanol–ethanol–water (40:11:19), he used a total of six different developer solvent systems. Apparently no one solvent system gave satisfactory results. Compounds were also separated by repeated preparative PC, but none were obtained analytically pure by this method.

More recently SCHMIDT<sup>12</sup> used PC for the fractionation of lignin sulfonates from spent sulfite liquors. Two separate procedures are described. Good separation of material is claimed when the following three irrigating solvent systems are used consecutively: 1-butanol–ethanol–water (25 % aqueous solution), 1-propanol–water (2:1), and 1-propanol–water (3:2). This procedure, used primarily for investigation of lignin sulfonates obtained from neutral sulfite semichemical spent liquors, would be time consuming considering three chromatographic development periods plus three drying times. For reasons not mentioned another solvent system was developed for comparing the lignin sulfonates from various sources. Here the solvent system employed was 1-propanol–ethanol–1.6 *N* ammonium carbonate solution–water (100:

532:268:100) with the relative amount of water varied as needed for maximum differentiation among the spent liquor samples being evaluated. SCHMIDT analyzed the chromatographic fractions by elution of the material from the paper and determination of UV absorption at 280 nm.

In the paper by SCHMIDT no  $R_F$  values are given nor were model compounds used as reference substances. The assumption is made that the higher the  $R_F$  the lower the molecular weight of the material. This assumption is probably valid for substances having the same approximate sulfonate (or sulfur) content. As will be discussed later in the text, a high sulfonate content results in low  $R_F$  values.

### Discussion

By use of a developer solvent system consisting of 2-butanone, acetic acid and water, low-molecular-weight lignin sulfonates can be effectively separated by means of PC. In the new solvent system proposed the primary influencing group of the lignin sulfonates is the highly ionized sulfonate group, which masks to a large extent any effect other groups within the molecule may exert. All compounds having one sulfonate group will be in one  $R_F$  range, and those compounds having two sulfonate groups will be in another, lower,  $R_F$  range. This phenomenon, although not as noticeable, was also obtained by GLENNIE<sup>10</sup> and PARRISH<sup>3,4</sup> in the solvent systems they employed. The size of the remainder of the molecule usually determines its position within an  $R_F$  range, with the larger this remainder the greater the  $R_F$  value. As a result, low-molecular-weight lignin sulfonates are grouped in the order of increasing  $R_F$  values as follows: monomeric disulfonates, dimeric disulfonates, monomeric monosulfonates and dimeric monosulfonates.

Through changing the concentration of the 2-butanone in the developer solvent system either monosulfonates, disulfonates, or both may be separated by PC. When an aqueous solution of 85% 2-butanone containing 5% acetic acid is used the monosulfonates will separate on the paper chromatogram and the disulfonates will remain at the origin. The  $R_F$  values are all low and it is necessary to allow the solvent front to extend beyond the paper. With an aqueous solution of 70% 2-butanone and 5% acetic acid the disulfonates will separate sufficiently on the chromatogram and the monosulfonates will all have high  $R_F$  values and be poorly separated. For the PC of low-molecular-weight lignin sulfonates from spent sulfite liquors a developer solvent system of 2-butanone-acetic acid-water (16:1:3), an 80% 2-butanone solution, was considered the best.

In Fig. 1 is shown a typical chromatogram, using Whatman No. 1 paper, of an isolated low-molecular-weight fraction of lignin sulfonate material and a composite chromatogram of lignin-type sulfonate model compounds. The source of the lignin sulfonate material was "Lignosite", a commercial spent sulfite liquor, resulting from the pulping of softwoods, available from the Georgia-Pacific Corporation, Bellingham, Washington. The low-molecular-weight fraction was obtained by fractionation of the quaternary ammonium salts of lignin sulfonates using essentially the procedure of QUIMBY AND GOLDSCHMID<sup>13</sup>. The lignin sulfonates were recovered as sodium salts by use of a Dowex 50 cation-exchange column. Preparation of the lignin-type sulfonate model compounds is given by GLENNIE<sup>10</sup>, SCHUBERT *et al.*<sup>6</sup> and JANSON AND SJÖSTRÖM<sup>14</sup>. The formulae of the model compounds and  $R_F$  values are given in Table I. In developing the chromatogram the solvent front

was allowed to extend considerably beyond the limits of the paper in order to separate the material in the low  $R_F$  range. As a consequence the  $R_F$  values given were calculated using the following procedure: The actual  $R_F$  of Reference No. 6 was obtained independently,  $R_F$  values were then determined for the other compounds and the  $R_F$  of Reference No. 6 used as a multiplication factor. The spots on the paper chromatograms were visualized by viewing under UV light with and without simultaneous ammonia fuming and by spraying with a diazo salt of 2-amino-4-nitroanisole (Fast Scarlet R Salt from Koppers Co., Inc.) followed by 0.1 *N* sodium hydroxide.

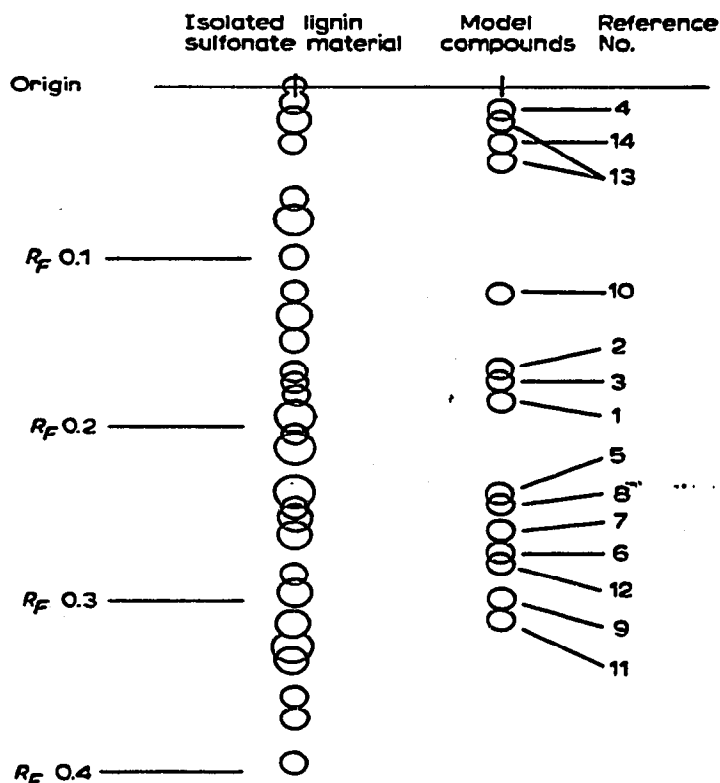


Fig. 1. Typical paper chromatogram of low-molecular-weight lignin sulfonate material and a composite chromatogram of lignin sulfonate-type model compounds. Irrigating solvent system: 2-butanone-acetic acid-water (16:1:3). Model compounds are listed by reference number in Table I.

In the 80% 2-butanone solvent system lignin-type monosulfonate model compounds have  $R_F$  values from 0.32 to 0.16 with the exception of one substance, containing a 1,2-glycol group with an  $R_F$  of 0.12. Both PARRISH<sup>8</sup> and GLENNIE<sup>10</sup> obtained a lower  $R_F$  value for similar compounds with a 1,2-glycol group. The dimer monosulfonate compounds had  $R_F$  values of 0.27 and 0.32 while monomer and dimer disulfonate compounds had  $R_F$  values less than 0.05.

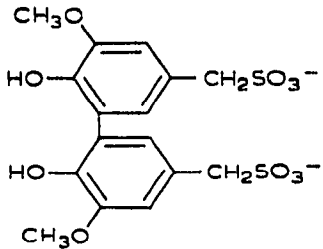
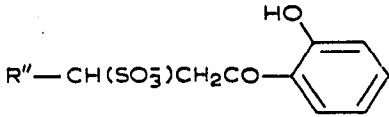
An exception to the above discussion regarding  $R_F$  ranges occurs when the *para*-hydroxy position is etherified, such as with a methyl substitution. The  $R_F$  range is considerably higher in lignin-type sulfonate compounds having a methoxy group in the *para* position instead of a free phenolic group. This is indicated by comparing reference compounds 15 and 16 in Table I with other related compounds. Although these sulfonated methoxy derivatives could occur in studies involving

the sulfite pulping of model compounds, they would not be present in low-molecular-weight lignin sulfonate fractions from natural sources. Only a free *para*-phenolic group would occur in monomeric and dimeric lignin sulfonates from the sulfite pulping of lignified material.

TABLE I

LIGNIN SULFONATE TYPE MODEL COMPOUNDS AND THEIR  $R_F$  VALUES

Solvent system: 2-butanone-acetic acid-water (16:1:3).

Reference No.	Formula <sup>a</sup>	$R_F$ value
1	$R-CH_2SO_3^-$ <sup>b</sup>	0.18
2	$R'-CH_2SO_3^-$	0.17
3	$R'-CH = CHCH(OH)SO_3^-$	0.17
4	$R'-CH(SO_3^-)CH_2CH(OH)SO_3^-$	0.02
5	$R'-COCH(SO_3^-)CH_3$	0.24
6	$R'-CH(SO_3^-)CH = CH_2$	0.27
7	$R'-CH = CHCH_2SO_3^-$	0.26
8	$R-CH(SO_3^-)CH = CH_2$	0.24
9	$R'-CH(OH)CH(SO_3^-)CH_3$	0.30
10	$R'-CH(SO_3^-)CH(OH)CH_2OH$	0.12
11	$R'-COCH_2CH(SO_3^-)-R'$	0.31
12	$R'-CH(OH)CH(SO_3^-)-R'$	0.27
13	$R'-CH(SO_3^-)CH(SO_3^-)-R'$	{ 0.02 0.04
14		0.03
15	$R''-COCH_2SO_3^-$	0.51
16		0.67

<sup>a</sup> R = *p*-Hydroxybenzyl; R' = guaiacyl; R'' = 4-methylguaiacyl.

<sup>b</sup> Cation not indicated, usually sodium.

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