Acknowledgements—The authors thank Prof. S. M. Sircar, Dr. D. M. Bose and Dr. A. Sen for their interest in the work. Thanks are also due to Dr. R. RAO, N.I.H. Bethesda, U.S.A. for the N.M.R. spectrum. The financial support to B.K.C. from the Department of Atomic Energy, India is gratefully acknowledged.

Phytochemistry, 1971, Vol. 10, pp. 483 to 484. Pergamon Press. Printed in England.

SALICACEAE

HOT WATER PHENOLIC EXTRACTIVES OF THE BARK AND LEAVES OF DIPLOID *POPULUS TREMULOIDES**

IRWIN A. PEARL and STEPHEN F. DARLING

The Institute of Paper Chemistry, Appleton, Wisconsin 54911, U.S.A.

(*Received* 24 *April* 1970)

Abstract—The hot water extractives of the fresh smooth green bark, leaves, and leaf stem twigs of a diploid *Populus tremuloides* tree cut in June were fractionated by ethyl acetate extraction and polyamide chromatography. Crystalline components isolated in quantity included salicin, salicortin, 1-*O*-*p*-coumaroylglucose, tremuloidin, tremulacin, and salireposide.

IN THE course of our studies on the barks and leaves of *Populus* species, the continuous evolutionary changes in processing employed in the laboratory suggested that organs of species investigated more than a decade ago be reinvestigated in the light of the newer knowledge. Accordingly, the smooth green bark, leaves, and leaf stem twigs of a diploid *P. tremuloides* tree cut in June were studied.

The fresh organs were processed immediately by the Waring Blendor and gradient elution polyamide chromatography procedures as detailed in an earlier paper on *Salix purpurea* bark.[1] The crystalline components isolated are presented in Table 1.

The recovery of crystalline products as noted in Table 1 represented a comparatively small proportion of the total products recovered from the chromatograms. TLC of filtrates from the crystals noted in the table in all instances indicated more of the specific compound together with one or more components, some in substantial amount. Further fractionation is necessary to separate these components in order to obtain crystalline compounds for identification. This is especially true in the case of compounds which are exceedingly soluble in water as represented by salicortin in the present study. The tremulacin noted in Table 1 was obtained by silica gel column chromatography as described earlier for salicortin.[1]

* Part XXVII in the series "Studies on the Barks of the Family Salicaceae" and Part XIV in the series "Studies on the Leaves of the Family Salicaceae."

[1] I. A. PEARL and S. F. DARLING, *Phytochem.* 9, 1277 (1970).

TABLE 1. CRYSTALLINE COMPONENTS FROM CHROMATOGRAPHY OF ETHYL ACETATE-SOLUBLE PORTION OF HOT WATER EXTRACTIVES OF DIPLOID *Populus tremuloides* ORGANS

Component	Bark yield, %*	Leaves yield, %	Twigs yield, %
Crude extract	8·55	5·94	11·45
Salicin	0·11	P	1·42
Mixed salicin and salicortin			0·52
Salicortin	0·59	P	0·84
Pyrocatechol	P	P	P
1-*O*-*p*-Coumaroylglucose	0·11		0·12
Tremuloidin	0·04	0·05	1·92
Mixed tremuloidin and tremulacin			0·27
Tremulacin	0·29	P	
Salireposide	0·27		
Rutin		0·04	
Flavonoid compounds	P	0·02	0·01
Total solids recovered from eluate	8·06	4·95	10·75

* On basis of oven-dry organ solids.
P = present in quantity by TLC, but not isolated and weighed.

In the elution curves of this study, as in those of an earlier study of triploid *P. tremuloides* bark,[2] a substantial portion of the total eluate solids was collected in the first ten eluate fractions. TLC of these fractions indicated essentially only salicin and salicortin, and these could be separated by silica gel chromatography.[1] In the case of the leaves study of Table 1, where no crystalline salicin or salicortin are noted, the two components alone amounted to 35 per cent of the total ethyl acetate-soluble extractives.

As in a previous study on the twig bark and trunk bark of *P. balsamifera*,[3] a comparison of the data of columns 1 and 3 of Table 1 demonstrate that the content of ethyl acetate-soluble water extractives of the twig bark is much higher than that of the trunk and branch bark of the same diploid *P. tremuloides* tree and, except for tremulacin, the twig bark is much richer in isolatable crystalline material than is the trunk and branch bark. The difference is even more pronounced because of the fact that the twigs employed in this study contained considerable xylem material besides the bark. The twigs of diploid *P. tremuloides* appear to be an excellent raw material for the production of tremuloidin.

EXPERIMENTAL

The smooth green bark, leaves, and leaf stem twigs of a diploid *P. tremuloides* cut in Outagamie County, Wisconsin in June were processed immediately exactly as described previously for *S. purpurea* bark.[1]

Acknowledgement—The authors wish to thank Mrs. Charlotte Robbins for her help in the processing and analytical monitoring required in this investigation.

[2] I. A. PEARL and S. F. DARLING, *Tappi* **50**, 324 (1967).
[3] I. A. PEARL and S. F. DARLING, *Phytochem.* **7**, 1855 (1968).