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[54] **SOLUBILIZATION OF PROTEINS FOR PHARMACEUTICAL COMPOSITIONS USING HEPARIN FRAGMENTS**

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[58] **Field of Search** 514/2, 8, 56; 424/85–87; 530/351, 402, 403, 405, 395, 389–391; 435/811

[56] **References Cited**

U.S. PATENT DOCUMENTS

3,619,371	11/1971	Crook et al. .	
4,055,635	10/1977	Green et al. .	
4,496,689	1/1985	Mitra	514/59
4,518,584	5/1985	Mark et al.	530/351
4,588,585	5/1986	Mark et al.	530/351
4,613,665	9/1986	Larm	525/34.2

FOREIGN PATENT DOCUMENTS

0154316	9/1985	European Pat. Off. .
0147761	7/1985	European Pat. Off. .
8603318	3/1985	PCT Int'l Appl. .
8700056	1/1987	PCT Int'l Appl. .

OTHER PUBLICATIONS

Kim et al., CA, vol 102, 1985, #219467r.

Function of Proteoglycans, ed. Evered et al., 1986, pages vary.

Kim et al., *Korean Biochem. J.,* 1985, vol. 18(1), pp. 50–55.

Hoffman et al., *Carbohydrate Research,* 117, 328–331, (1983).

Hoylaerts et al., *Thromb. Haemostas* (Stuttgart) 49:109 (1983).

Ceustermans et al., *J. Biol. Chem.,* 257:3401 (1982).

Bjork et al., *FEBS Letters,* 143(a), 96 (1982).

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[57] **ABSTRACT**

A pharmaceutical composition is prepared wherein a biologically active conjugated protein which is β-interferon, interleukin-2, or an immunotoxin is dissolved in an aqueous carrier medium without the presence of a solubilizing agent. The unconjugated protein, which is poorly or not at all water-soluble at pH 6–8 without such solubilizing agent, is selectively conjugated to at least one heparin fragment having a terminal 2,5-anhydro-D-mannose residue which has an aldehyde not involved in intramolecular hemiacetal formation.

16 Claims, No Drawings