RESEARCH PAPER

Accepted 7 November 2003
Revised 28 August 2003
Received 10 February 2003
Indian J. Pharm. Sci., 2004, 66(1): 69-71

Coupling of Indomethacin to Poly (HEMA); *In Vitro* and Bioavailability Characterization

M. J. N. CHANDRASEKAR*, M. J. NANJAN AND B. SURESH
Department of Pharmaceutical Chemistry
J. S. S. College of Pharmacy, Ootacamund-643001.

A polymeric pro-drug of indomethacin was synthesised. The pro-drug was evaluated for drug content and *in vitro* drug release behavior at pH 1.2 and 7.2. The *in vitro* and studies show that the drug-release takes place predominantly at the higher pH and also in a sustained manner as hypothesized. The bioavialability investigations show complete drug absorption from the polymeric pro-drug, thus showing its potential for site-specific and sustained drug delivery.

NSAIDs, though generally effective in the management of pain and inflammation, are also associated with the development of several gastrointestinal complications especially in stomach. Direct damage to the lower GI tract is, however, unusual[1]. Several approaches have been made to overcome the GI complications like cotherapies with sucralfate[2], H_2 antagonists[3] and prostaglandin inhibitors[5] have been tried. In recent years NSAIDs based on COX-2 selective compounds have also been used[5]. All these approaches, however, have not been able to provide the required complete protection against GI complications.

In recent years, polymeric drug derivatives in which the drug molecules are linked to polymeric matrices through covalent bonding of limited stability in physiological environment, is receiving considerable attention[6-11]. This is believed to be one of the most promising ways to modify the pharmacokinetics of the drugs and to achieve preferential localization to target sites.

It was proposed, therefore, adopt a polymeric pro-drug approach wherein indomethacin is covalently attached to poly (hydroxyethyl methacrylate) [poly (HEMA)] a biocompatibile[8] polymer through an ester linkage. Such a system is expected to preferentially cleave and release the drug in the alkaline environment of the lower GI tract in a site-specific manner, rather than the acidic environment of the upper GI tract. We report here the synthesis, the *in vitro* drug release behavior at different pH and bioavialability of the polymeric pro-drug.

*For correspondence
E-mail: ncsekar_in@yahoo.com

MATERIALS AND METHODS

2-Hydroxyethyl methacrylate (HEMA) was obtained from M/s Fluka, Switzerland. Benzoyl peroxide was obtained from Wilson Laboratory, Switzerland; thionyl chloride was obtained from S. D. Fine Chemicals, Mumbai. Indomethacin was a gift sample from Tablets India, Chennai. Permission has been obtained from the Institutional Animal Ethics Committee (IAEC) to carry out the *in vivo* experiments, (CPCSEA/CH/CRG/2001/OOTY).

Preparation of the polymeric pro-drug:

HEMA (26.4 g) was taken in a 250 ml three-necked round bottom flask fitted with a stirrer and condenser. Thionyl chloride (26.2 g) was added drop wise and the reaction was carried at 70° for 3 h. Chloro ethyl methacrylate obtained as a liquid was purified by distillation. The monomeric drug derivative was prepared by reacting sodium salt of indomethacin (5 g) with chloroethyl methacrylate (3.7 g) in DMSO at 120° for 8 h. The contents of the flask were poured into distilled water when a precipitate was formed. It was filtered, dried and purified. Its IR spectrum was recorded using Perkin-Elmer 1600 series FT-IR spectrophotometer. To (5 g) of the monomeric drug derivative in DMSO benzoyl peroxide (0.1 g) was added to initiate polymerization. After 6 h the contents of the flask were poured into 500 ml of

distilled water. The precipitate of polymeric pro-drug obtained was filtered and dried and purified.

Estimation of drug content:

The drug content was estimated spectrophotometrically by first treating the pro-drug (100 mg) with 0.1 M NaOH (100 ml) to achieve complete release of the drug from the polymeric backbone through hydrolysis.

***In vitro* drug release:**

The *in vitro* drug release a study of the polymeric pro-drug was carried out at two different pH levels, pH 1.2 and pH 7.2, which mimic the pH in upper and lower GI tract over a period of 24 h. Samples were withdrawn at different time intervals and analysed spectrophotometrically.

Bioavialability studies:

A comparative bioavialability study was carried out in New Zealand rabbits. Free drug and pro-drug containing an equivalent quantity of indomethacin were administered orally as suspension in 0.5% w/v carboxy methylcellulose. Blood samples were withdrawn at different time intervals over a period of 24 h and a reversed-phase HPLC method[12] was adopted to quantitate the drug-plasma levels.

RESULTS AND DISCUSSION

The scheme of synthesis of the polymeric pro-drug is represented in fig 1. The IR spectrum of the monomeric drug derivative exhibits intense bands at 1734 cm^{-1} (C=O) and 1653 cm^{-1} (C=C), 1016 cm^{-1} (C-O-C) and the absence of a band at 760 cm^{-1} (C-Cl). H^1 NMR in $CDCl_3$, δ: 2.38 (3H, S, -CH_3), 3.7 (2H, S, -NCH_2-), 3.82 (3H, S, -OCH_3), 4.01 (5H, M, CH_3-C=C-, -CH_2COO-), 6.82 (4H, S, aromatic ring with Cl atom), 7.62 (5H, M, aromatic ring). The drug content was estimated to be 0.712 mg/g of the polymeric pro-drug.

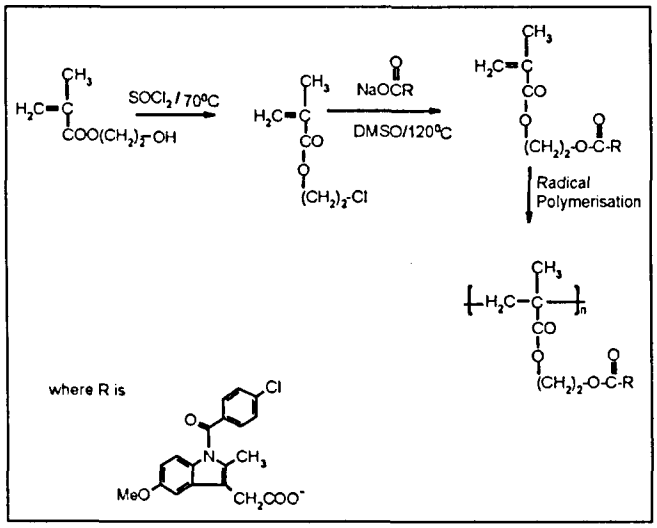

Fig.1: Synthesis of the polymeric pro-drug.

The *in vitro* drug release profile of the pro-drug synthesised is given in fig. 2. At pH 7.2 a burst release of 33% was observed within 1 h followed by a sustained release over a period of 12 h. A maximum of 94% of the drug was released from the pro-drug. Though there is a sustained drug release at pH 1.2, the amount of drug released at various time intervals was comparatively lower than that at pH 7.2. The slow drug release at pH 1.2 is due to fact that hydrolysis reaction in an acidic environment is reversible whereas in an alkaline environment hydrolysis reactions are irreversible reactions. In other words, hydrolysis of esters in the alkaline environment is expected to proceed to completion[13]. The polymeric pro-drug synthesised thus releases the drug predominantly in the alkaline environment in a site-specific manner.

The bioavialability study showed detectable concentrations of the drug till 18 h in the case of pro-drug but only for 10 h for the free drug. The peak plasma concentration for the free drug was 601 ng/ml within 1 h (T_{max}) whereas for the pro-drug it was 223 ng/ml after a period

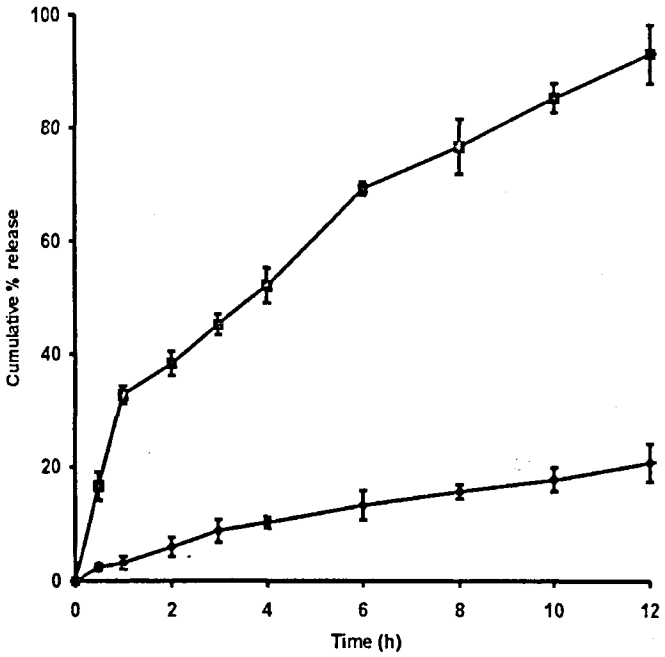

Fig.2: *In vitro* drug release profile of the pro-drug.

In vitro drug release profile of the pro-drug carried out using USP XXXIII dissolution apparatus at pH 1.2 (-□-) and pH 7.2 (-■-).

of 6 h. The delay in T_{max} is thus due to the slower rate of drug release from the pro-drug and its consequent absorption. There was no significant change in the extent of drug absorption between the free drug and the pro-drug as the area under the plasma concentration-time curve (AUC) is almost the same, namely, 2143 and 2311 ng h/ml, respectively. This confirms the complete drug release from the polymeric pro-drug and its potential in sustaining the drug release under *in vivo* conditions.

In summary, the results indicate that the covalent linkage of indomethacin to a biocompatible polymer, poly(HEMA), through an ester group leads to a delivery system, which is capable of releasing the drug in a sustained and site-specific manner to the lower gastrointestinal tract.

ACKNOWLEDGEMENTS

The authors would like to thank Sri Jagadguru Sri Shivarathreeswara Deshikendra Mahaswamiji of Suttur Mutt for providing the facilities to carry out the work.

REFERENCES

1. Peterson, S.P. and Killurman, I., **Postgraduate Med.,** 1996, 100, 131.
2. Agarwal, N. M., Roth, S. and Graham, D.Y., **Ann. Intern. Med.,** 1991,115, 195.
3. Robinson, M.G., Griffn, J.W. and Bowers, J., **Dig. Dis. Sci.,** 1989, 34, 424.
4. Graham, D.Y., White, R.H. and Moreland, L.W., **Ann. Intern. Med.,** 1993, 119, 257.
5. Roth, S.H., Tindall, F.A. and Jain, A.K., **Arch. Intern. Med.,** 1993, 153, 2565.
6. Hawkey, C.J., **Gastroenterology,** 1995, 109, 614.
7. Gershonov, E., Gold waser, I., Fridkin, M. and Sheehter, Y., **J. Med. Chem.,** 2000, 43, 2530.
8. Battaghia, G., La Russa, M., Bruno, V., Aranare, L., Ippolito, R., Copani, A., Bomina F. and Nicoletti, F., **Brain Res.,** 2000, 860, 149.
9. Rantio, J., Nevalainen, T., Taipale, H., Vepsalainen, J., Gynther, J., Laire K., and Jarvinen, T., **J. Med. Chem.,** 2000, 43, 1489.
10. Gac, S., Condane, J., Boustta, M., Domurado, M., and Vert, M., **J. Drug Targeting,** 2000, 7, 393.
11. Hashida, M., Akarnaslu, K., Nishikawa, M., Yarnashita, F., Yoshirawa H., and Takakura, Y., **Pharmazie,** 2000, 55, 202.
12. Mason, J.L. and Hobbs, G.J., **J. Liq. Chromatogr.,** 1995, 8, 2045.
13. Stanley, H.P., In; Organic Chemistry, 5th Edn., McGraw Hill International Edition, New York, 1987, 278.