Starch is a carbohydrate consisting of a large number of glucose units joined together by glycosidic bonds. It consists of two types of molecules: the linear and helical amylose and the branched amylopectin. Depending on the plant, starch generally contains 20 to 25% amylose and 75 to 80% amylopectin [10]. Porous starch is a biodegradable starch that has great potential as a solid dispersion carrier for oral poorly water soluble drugs. Porous starch has a nano-porous structure, low density, high specific surface area and pore volume; its distinctive advantages include no toxicity, biocompatibility, and biodegradability [11]. Traditionally porous starch has been prepared by swelling or by heat assisted microwave technique [12].

Diclofenac is a non-steroidal anti-inflammatory drug (NSAID) and is poorly water insoluble. It is widely used to treat swelling and pain in rheumatoid arthritis, osteoarthritis and ankylosing spondylitis.

This work was aimed at exploring the possibility of improving solubility and dissolution rate of poorly water soluble drug Diclofenac using the prepared porous starch as solid dispersion carrier

MATERIALS AND METHODS

Chemicals and Reagents

Diclofenac was obtained as gift sample from Blue Cross Laboratories Limited, Starch, Methanol was purchased from SD Fine Chem Pvt. Ltd, Mumbai, India. All other analytical grade reagents were obtained commercially and used as received. Double distilled water was used throughout the work.

Preparation of starch phosphate

Starch phosphate was prepared based on the method as described by Choi et al (18) with some modifications. Potato starch (10g) and di-sodium hydrogen orthophosphate anhydrous (3g) were suspended in 10 ml of water and continuously stirred for 20 min. This starch slurry was then filtered and the wet starch mixture was conditioned for 12 h at room temperature. To enhance phosphorylation, this mixture was heated in a forced air oven at 130 °C for 3 h. The product obtained was grounded and sized.

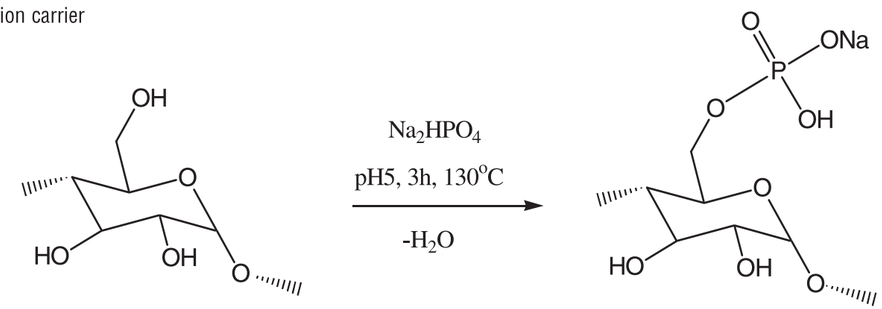

Figure 1: Phosphorification of Potato Starch to Produce Starch Phosphate

Preparation of solid dispersions of Diclofenac sodium in starch phosphate

Solid dispersions of diclofenac and starch phosphate were prepared in 1:1 (SD-1), 1:2 (SD-2) & 1:3 (SD-3) weight ratios of drug: carrier by solvent evaporation method. Diclofenac sodium (1 g) was dissolved in methanol (5 ml) in a dry mortar to get a clear solution. Starch phosphate (1g) was then added and mixed. The thick slurry was triturated for 15 min for complete evaporation of dichloromethane and then dried at 55 °C until dry. The dried mass was pulverized and sieved through mesh no. 72 and stored in desiccators till further use.

1. Preparation of Diclofenac tablets

The tablets are prepared by conventional wet granulation method. Prepared SD-2 and SD-3 previously passed through #60 mesh screen were mixed in a stainless steel bowl. The powder blend was moistened with the required amount of water and then granulated using #18 mesh screens. The granules were dried at 60 °C for sufficient period of time and the moisture content of the granules as measured using the IR Moisture Meter was found to be confined within 0.85% to 1.2%. The dried granules were then passed through #22 mesh screen, mixed with magnesium stearate, and compressed into tablet using a flat-faced 10-mm punch in a ten-station rotary minipress tablet machine (RIMEK, Karnavati Engineering, Gujarat, India).

2. Characterization of starch phosphate

The starch phosphate prepared was evaluated for following parameters:

2.1. Solubility

Solubility of starch phosphate was tested in water, aqueous buffers of pH 6.8 and organic solvents such as alcohol, chloroform, and acetone (23).

2.2. pH

The pH of a 1% w/v slurry was prepared and pH is measured using pH meter (Systonic Limited, Kolkata).

2.3. Melting point

Melting point was determined by using melting point apparatus (24).

2.4. Swelling index

Starch phosphate (200 mg) was added to 10 ml of water and light liquid paraffin was taken in two different graduated test tubes and mixed. The dispersion in the tubes was allowed to stand for 12 h. The volumes of the sediment in the tubes were recorded. The swelling index of the material was calculated as follows.

$$S.I.(\%) = \left(\frac{\text{Volume of sediment in water} - \text{Volume of sediment in light liquid paraffin)}}{\text{Volume of sediment in light liquid Paraffin}}\right) \times 100$$

2.5. Bulk density

Bulk density (g/cc) was determined by funnel tap method in a graduated cylinder (24).

2.6. Angle of repose

Angle of repose was measured by fixed funnel method. For determination of angle of repose (è), the sample was poured through the walls of a funnel, which was fixed at a position such that its lower tip was at a height of exactly 2.0cm above hard surface. The sample was poured till the time when upper tip of the pile surface touched the lower tip of the funnel. The $\tan^{-1}$ of the (height of the pile / radius of its base) gave the angle of repose [25] .

2.7. Compressibility index

Compressibility index (CI) was determined by measuring the initial volume (V_o) and final volume (V) after hundred tapping of a sample of starch phosphate in a measuring cylinder. CI was calculated using equation(24).

$$\text{Compressibility index (CI)} = \frac{V_o - V}{V_o} \times 100$$

2.8. Physical characterisation of tablets

Weight variation test (using Precisa Electronic Balance, model XB 600M/C, Switzerland), diameter measurement (using Digimatic Caliper, model CD-6"CS, Mitutoyo Corporation, Japan), hardness test (using Monsanto type hardness tester), and friability test (using Friabilator, Veego, Mumbai, India) were done following the usual methods.

2.9. *In vitro* release study

In vitro drug release study was carried out in acidic solution 0.1 (N) HCl (pH 1.2) for 4 h and in USP PB solution (pH 6.8) using USP II dissolution rate test apparatus (model TDP-06P, Electro Lab, Mumbai, India). One weighed tablet was placed in 900 ml acidic solution (37± 1°C) and rotated with paddle at 75 rpm. Aliquot was withdrawn at different times and replenished immediately with the same volume of fresh solution. The withdrawn samples following suitable dilution were analyzed spectrophotometrically at 274 nm for acidic and buffer solution. The amounts of drug released in acidic medium and PB solution were calculated from the calibration curves drawn, respectively, in 0.1 (N) HCl and PB solution (pH 6.8). Each release study was duplicated.

RESULT and DISCUSSION

Starch phosphate was prepared by reacting starch with di-sodium hydrogen orthophosphate anhydrous at elevated temperatures. The reactions involved are shown in Fig. 1. Starch phosphate prepared was found to be white, crystalline, non hygroscopic powder and can easily be grounded to different sizes. Powder which passes through mesh no.72 and retained on mesh no.100 was collected. The starch phosphate prepared was characterised by determining various physical properties. The properties of starch phosphate prepared are summarised in Table 2. When tested for melting point, it was charred at 208 °C. Starch phosphate prepared was insoluble in water, aqueous fluids and several organic solvents. In water it exhibited good swelling (320%). No gelling/pasting was observed with starch phosphate when its aqueous dispersion was heated at 100°C for 30 min, where as potato starch formed a paste/gel during the above heat treatment. In the micromeritic evaluation, the angle of

repose and compressibility index values revealed the excellent flow characteristic of starch phosphate prepared (Table no 1).

As starch phosphate, a chemically modified starch was found to be insoluble in water and has good swelling property without pasting or gelling when heated in water it is considered as a promising carrier for solid dispersions for enhancing the dissolution rate of poorly soluble drugs. Solid dispersions of diclofenac in starch phosphate were prepared by solvent evaporation method employing various weight ratios of drug: starch phosphate.

For each designed formulation, a blend of drug and Starch phosphate was prepared and evaluated. Bulk density was found 0.3627±0.003 gm/cc and 0.374±0.004 gm/cc for 1:2 and 1:3 ratios respectively. For density data, Carr's index was calculated. Flowability of the material was found to be good as indicated by compressibility-flowability correlation data. Angle of repose was found in the range of 32.73±0.587° and 31.42±0.457° for 1:2 and 1:3 ratios respectively.

Tablets were prepared by wet granulation method. As the material was a free flowing, tablet were obtained of uniform weight due to uniform die fill, with acceptable variation as per I.P. specification (Table 3). Hardness of the tablet for each type formulation was 6.5 and 7.5 kg/cm^2. Friability was below than 1.0% shows an indication of good mechanical strength resistance of the tablet. The *in-vitro* disintegration time for solid dispersion tablet is less than 15 minutes (Table 3) The *in-vitro* drug release study indicated that more than 50% drug released in tablet with solid dispersion within 10 minutes whereas only 20% drug was released within 10 minutes from marketed preparation. Formulation with hydrophilic carrier, the dissolution rate of Diclofenac was increased at a significant level when compared other tablets. Many factors contributed to faster drug release rate such as decrease in particle size, decrease in agglomeration of particles, increase wettability and decrease in crystallinity of the drug and might be due to combined effect of improved wettability, emulsifying effect of carriers and reduction in particle size during the formation of solid dispersions. Overall increase in the dissolution performance of the optimized formulation was described in terms of dissolution parameters and when compared with pure drug, all the above parameters were increased in case of SD3 formulation. All these formulation also followed Hixson Crowell cube root dissolution equation this shows the formulation had better dissolution and dispersion characteristics.

CONCLUSION

Starch phosphate prepared by reacting starch with di-sodium hydrogen orthophosphate anhydrous at elevated temperatures was insoluble in water and has good swelling (320%) property without pasting or gelling when heated in water. Solid dispersions of diclofenac in starch phosphate prepared by solvent evaporation method employing various weight ratios of drug: starch phosphate gave rapid and higher dissolution of diclofenac when compared to pure drug. Dissolution followed first order kinetics. Solid dispersions of diclofenac prepared employing starch phosphate as carrier showed marked enhancement in the dissolution rate of diclofenac.

Table 1: Physical Properties of the Starch Phosphate Prepared

Property	Results
Solubility	Insoluble in aqueous and organic solvent
pH(1%w/v aqueous dispersion)	7.2
Melting Point	Charred at 208 xC
Swelling Index	320
Gelling property	No gelling properties and swollen particle are separated.
Bulk Density	0.445 gm/cc
Angle of Repose	22.16x
Compressibility Index	19.23%

Table 2: Evaluation of solid Dispersion 1:2 and 1:3 (Diclofenac: Starch phosphate)

Parameter	Pure Drug	Solid dispersion (1:2)	Solid dispersion (1:3)
Drug content	100	90.056±0.3275	99.52 ± 0.5241
Saturation solubility	Slightly	Very	Very
Bulk Density	0.4484±0.0018	0.3627±0.003	0.374±0.004
Carr's Index	15.52±0.267	23.54±0.265	20.15±0.249
Angle of Repose	29.19±0.187	32.73±0.587	31.42±0.457

Table 3: Evaluation of Diclofenac tablet

Parameter	SD 2	SD 3
Wt variation	Passes	Passes
Tablet Hardness (kg/cm2)	6.5	7.5
Friability (%)	0.9	0.7
In-Vitro DT (min)	07 (pass)	08 (pass)

Table 4: Dissolution Parameters of the Solid Dispersions of Diclofenac Prepared Employing Starch Phosphate as a Carrier

Formulation	PD15 (%)	T50 (min)
Diclofenac tablet(x)	2.76	>60
SD 2	53.86	<10
SD 3	61.15	<10

PD15 (%): percent dissolved in 15 min; T50 (min): time for 50 % dissolution;

Solid dispersion (1:2): Ratio of drug: starch phosphate in solid dispersions: SD-2(1:2);

Solid dispersion (1:3): Ratio of drug: starch phosphate in solid dispersions: SD-3(1:3).

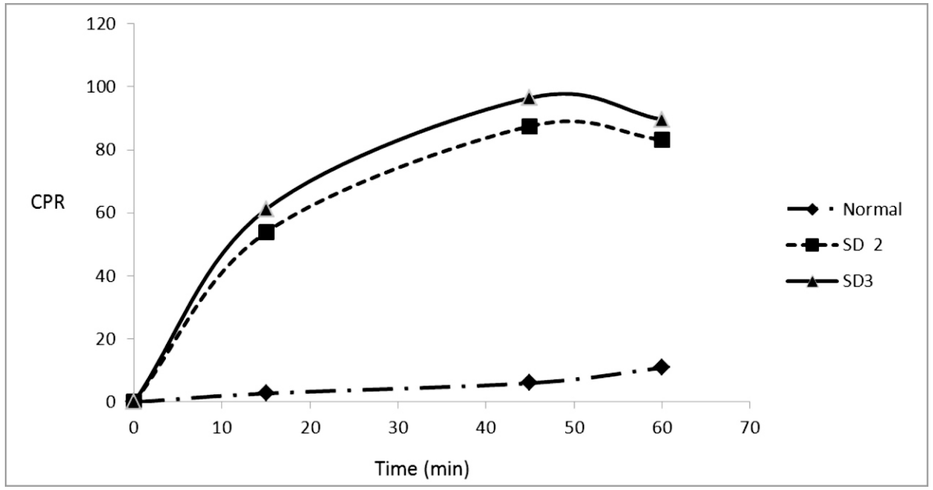

Figure 2: *In-vitro* drug release profile of Diclofenac tablet

The above picture describe the graphical representation of Cumulative percentage release (CPR) Vs. time of a pure drug, and drug containing solid dispersion in different ratio where in X and Y axis represents CPR and time (in minute) respectively.

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SYNTHESIS, CHARACTERIZATION AND EVALUATION OF ANTIMICROBIAL ACTIVITY OF 2, 4, 6-TRISUBSTITUTED PYRIMIDINE DERIVATIVES

Puspita Roy, Tanmoy Guria, Tanushree Singha, Benu Pani Sahu, Subbiah Ramasamy, Amalesh Samanta, & Tapan Kumar Maity*

Department of Pharmaceutical Technology, Synthetic and Natural Product Research Laboratory, Jadavpur University, Jadavpur, Kolkata-700032, West Bengal, India

*Corresponding author : Prof. Tapan Kumar Maity
e-mail : jutkmaity@yahoo.com, puspita.roy16@gmail.com, Mob.- 09432299486

ABSTRACT

In a wide search program toward new antimicrobial agents, we describe the synthesis of some pyrimidine derivatives (5a-d) from a series of chalcone analogues. These chalcone analogues were synthesized by condensation with substituted acetophenones and various aromatic aldehydes in ethanolic NaOH solution. The final compounds were characterized by using FT-IR and ^{1}HNMR spectroscopy. The antimicrobial activity of the novel products was evaluated against bacterial strains such as *Bacillus cereus, Bacillus subtilis, Micrococcus luteus, Pseudomonas aeruginosa* and *Bacillus pumilus*.

The result shows that drugs 5a and 5b have antimicrobial properties. Both the drugs were effective against five tested organisms among the listed organisms. Lowest Minimum Inhibitory Concentration of drug 5a and 5b was found at 200 µg/ml and the highest concentration was 400 µg/ml.

Keywords : Chalcone, Pyrimidine, MIC, Antimicrobial activity.

INTRODUCTION

At present, there is growing interest in the discovery of new antibacterial agents to bate against pathogenic microorganism, especially the bacteria resistant to the current antibiotics. Synthetic chemistry plays a key role in the development of new drug molecules for many years. With the advancement of modern chemistry and very sophisticated techniques it is has now become very easier to synthesize, characterize and evaluate new chemical entities (NCEs) [1].

Heterocycles form by far the largest of classical divisions of organic chemistry are of immense importance biologically and industrially [2].

Pyrimidines are 6-membered heterocyclic ring compounds composed of nitrogen and carbon. They are present throughout nature in various forms. Hundreds of pyrimidine-containing compounds have been found in biological system which control normal physiology. Pyrimidine ring is present in several pharmacologically active compounds, showing a wide range of biological activities, such as diuretic, anesthetic, anthelmintic, analgesic, anti-inflammatory etc [3, 4].

An antimicrobial is an agent that kills microorganisms or inhibits their growth. Antimicrobial medicines can be grouped according to the microorganisms they act primarily against [5, 6].

In an effort to develop potent anti-microbial agents, we have