Synthesis and Characterization of Chitosan Based Films Prepared from Narmada Riverside Crab Shells.

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Abstract
The present article describes the preparation and characterization of chitosan based films. The raw material was collected from Narmada riverside crab shells. The SEM images were taken for crab shell powder obtained from Narmada river side crab shells to observe the morphology. The chitin, chitosan and chitosan based films developed from the crab shell were also examined for morphology. The SEM photographs of crab shell powder, prepared chitin and chitosan exhibited rough and thick surface morphology under electron microscopic examination. The pure chitosan film demonstrated uniform and homogenous surface confirming the formation of an ordered film structure. Unlike the pure chitosan film, the surface of plasticized films became coarse and exhibited small cluster of particles attributed to glycerol presence. The XRD analysis of prepared chitosan films was performed. The pure chitosan film found to be amorphous in nature. Wide hump observed at 2θ=11.23 and 23.92. As a function of doping with different glycerol percentage it become crystallized.

Keywords- Chitin, Chitosan, Crab Shells, Narmada Riverside, and Chitosan based Films.

Introduction

Crab Shell is the natural composite material consisting of calcium carbonate, protein, and chitin. There is vast scope in mechanical studies of this hard shell. Crabs are covered by an exoskeleton, which is periodically shed as the animal grows. The exoskeleton of the crab consists mainly of chitin biopolymer. The calcium carbonate gives mechanical rigidity to the crab shell. The crab shell is multifunctional: it resists mechanical loads, supports the body, and provides environmental protection and resistance to desiccation [1-6]. In crab exoskeletons, the minerals are in the form of calcite or amorphous calcium carbonate, deposited within the chitin–protein matrix [7-9]. It is important to know the material nature whether crystalline or amorphous. X-ray diffraction analysis helps to understand crystallinity and amorphous nature of the materials.

Chitosan, like other natural polymers, is known to be partially crystalline polymer where the crystallinity is formed as a result of accumulation of linear chains. Chitin has been extracted from the crab shell and its mechanical strength investigated [10] Barbakadze et al. [11] investigated the mechanical properties for beetle. He worked on head cuticle of it. The outer shell of insects has great mechanical properties. They used SEM and TEM to study the structure of the shell..Verma and Tomar [12], studied the exoskeleton thermal behavior. They experimented the exoskeleton at high temperature. The study was made on shrimp shell with temperature ranging from 30ºC to 80ºC.Within the reported temperature range nanoindentation was carriedout.The SEM image showed the Bouligand pattern. This bouligand pattern is the main character of custanceans. Abdel-Rahmana et al. [13], Extracted chitin from shrimp shells. The shell waste was collected from Brazilian coast. They also prepared chitosan from the chitin and had characterization. The chemical method was used to extract chitin. The obtained chitin and chitosan was confirmed by XRD, ATR-FTR, SEM, H/C NMR, UV-Vis spectroscopy. Tijani et al. [14], extracted chitin from Nigerian sources. The investigation was done for extraction and characterization of the produced chitin. The chemical method was used to deproteinize and to demineralise the shells.XRD and SEM were performed. The uniform structure was observed in SEM. Ahemed et al. [15], worked on composite film. The chitosan and graphene oxide used to develop composite film. The various properties like mechanical behavior, thermal behavior, structural property and barrier property of the composite film were investigated.SEM and XRD were done for material characterization. Cesano et al.[16], investigated and synthesized the composite film. They produced the magnetic chitosan film. The surface magnetic behavior and bulk property were investigated. To understand the organic and inorganic properties of the film, one step method was used. The material composition was examined by XRD. The applications of the crab based chitin are described showing various uses in engineering and technology [17].

2. Materials and Methods

2.1 Crab shell Collection

The raw material used in present work is Narmada River Side Crab Shell collected from the fish markets of Nimad region of Khargone (MP) India. Crab shells were cleaned and washed thoroughly to remove any foreign materials, followed by grinding to get particle size 0.30-0.35 mm. The crab shell were collected and processed to extract chitin and chitosan. Grinding, demineralization, deproteinization etc. were the main steps followed by filtration and drying. The

chitin and chitosan extracted as described by Gadgey and Dey 2017[18] with some modifications.

2.2 Synthesis of Chitosan Based Films

In this study chitosan films were prepared by solution casting method. The prepared chitosan was used for film preparation. Acetic acid was used as a solvent and glycerol was used as a plasticizer. The films were prepared by the solution casting as described by Han et al.[19] with some modifications. The extracted chitosan from Narmada riverside crab shells were used to prepare the chitosan film. In a 250 ml beaker 100 ml distilled water was used to dissolve 2gm chitosan with 2 ml acetic acid. The solution was stirred overnight with magnetic stirrer on hot plate at 40ºC temperature. After complete dissolution of chitosan, glycerol was added as a plasticizer. Four moulds were prepared with 0%, 30%, 60% and 90% glycerol (w/w) and the prepared films were coded as CS-G00, CS-G30, CS-G60, and CS-G90 respectively. The solution was filtered through coarse sintered glass filter to remove un dissolved impurities. The solution was poured onto glass moulds. The films were dried for four days at ambient temperature. The films were peeled off and conditioned for testing. The peeled off films were cut into pieces to prepare specimens. The specimens were stored separately according to their compositions. The mechanical properties of chitosan based films were investigated previously [20.21].Thermal investigation of chitin was also done [22].

2.3 Characterization of Chitosan Based Films

2.3.1 Scanning Electron Microscopy (SEM)

The specimen are non conductive, hence to make them conductive gold coating of specimen is required. This process was carried out at UGC-DAE Consortium for Scientific Research, DAVV campus Indore (MP) India. The gold coating machine Quorum Q150TS used in the present study. The gold coating of 25 A° was done. The coating thickness was taken only 25 A° because more thickness could hide the original surface morphology.

The SEM images were taken at UGC-DAE Consortium for Scientific Research, DAVV campus Indore (MP) India. The Scanning Electron Microscope, model JEOL JSM 5600 was used. The SEM images were taken for crab shell powder, chitin, chitosan, and chitosan based films with different proportion of Glycerol.

2.3.2 X-ray Diffraction (XRD)

The X-ray diffraction analysis is done at UGC-DAE Consortium for Scientific Research, DAVV campus Indore (MP). The chitosan based films were analyzed on Bruker D8 Advance XRD machine(Germany) with Theatha-2Theata Geometry, with Ni-filtered Cu Kα radiation at voltage of 40 kV and current of 40 mA (λ = 0.154 nm).

3. Results and Discussion

3.1 SEM Analysis

SEM surface micrographs of Narmada Riverside Crab Shell powder, Chitin powder, Chitosan powder, pure chitosan film (CS-G00), plasticized chitosan films, CS-G30, CS-G60 and CS-G90 films are illustrated in Figures 1-7. The SEM photographs of crab shell powder, chitin and chitosan exhibited rough and thick surface morphology under electron microscopic examination. The films produced with the highest proportion of glycerol presented certain structural discontinuities and roughness along the entire surface, as well as small solid particles, contrarily to that present in films with pure chitosan, which had a smooth and very homogeneous surface. The pure chitosan film (CS-G00) shows smoothest, compact, uniform and homogenous surface confirming the formation of an ordered film structure. Unlike the pure chitosan film, the surface of plasticized films became coarse and exhibited small cluster of particles attributed to glycerol presence. The extent of roughness and clusters of particles increased with increasing the plasticizer loading. The SEM image of plasticized films reveals scanty clusters; these clusters are the protruding glycerol films which are entrapped and thickly coated with chitosan. Similar type of structures were also observed by Pan et al. 2011[23] for chitosan - graphene oxide composite films. Similar SEM images are also reported by Ahmed et al. 2017[15]. Baron et al. 2017[24] also observed the similar results for pure chitosan film surface morphology.

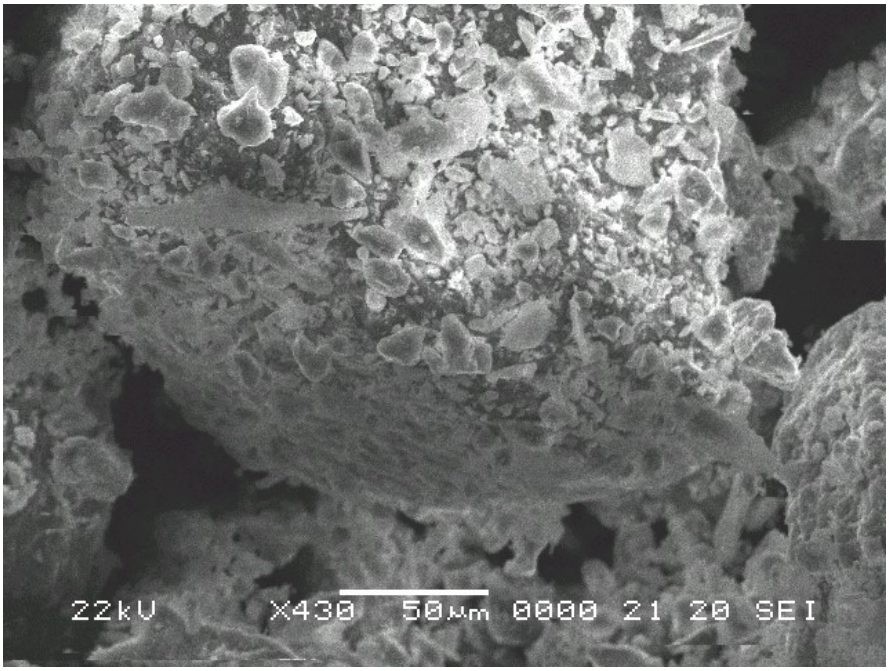

Figure 1 **SEM** micrographs of **Crab shell Powder**

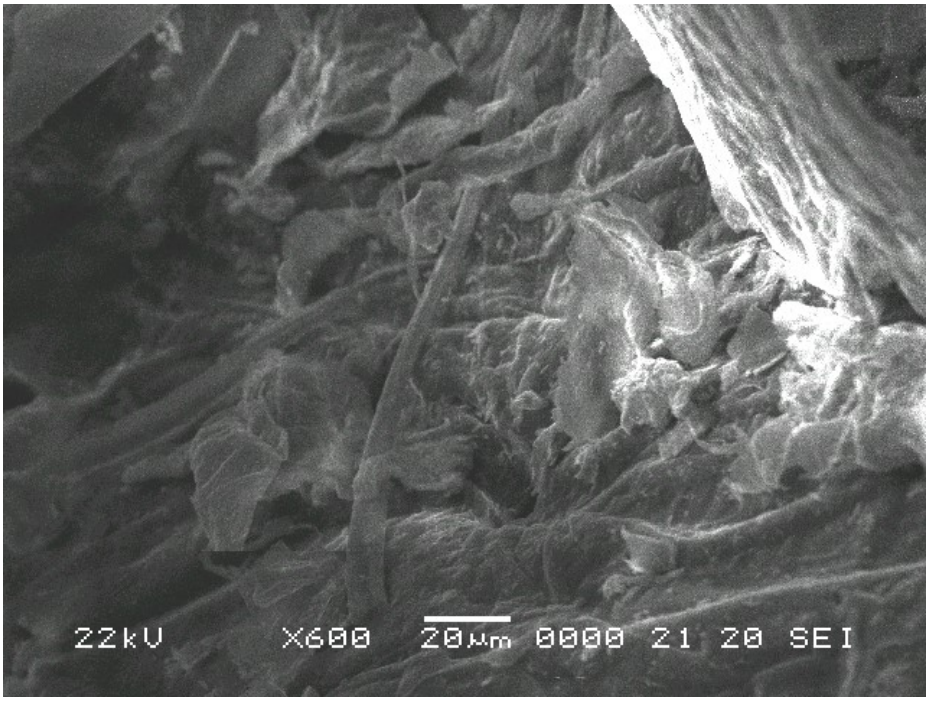

Figure 2 **SEM** micrographs of Chitin Powder

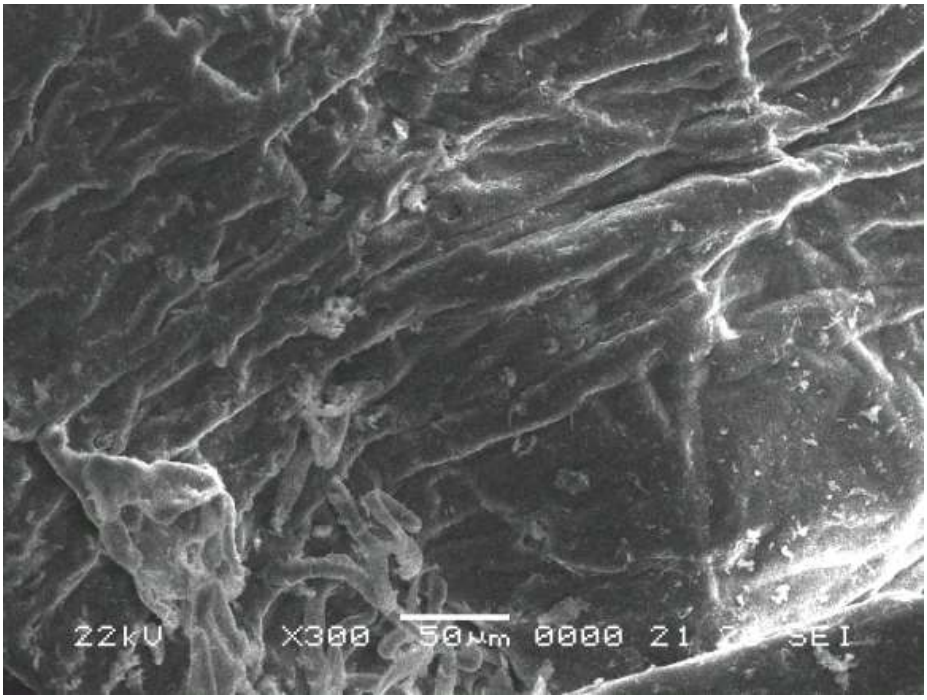

Figure 3 **SEM** micrograph of **Chitosan Powder**

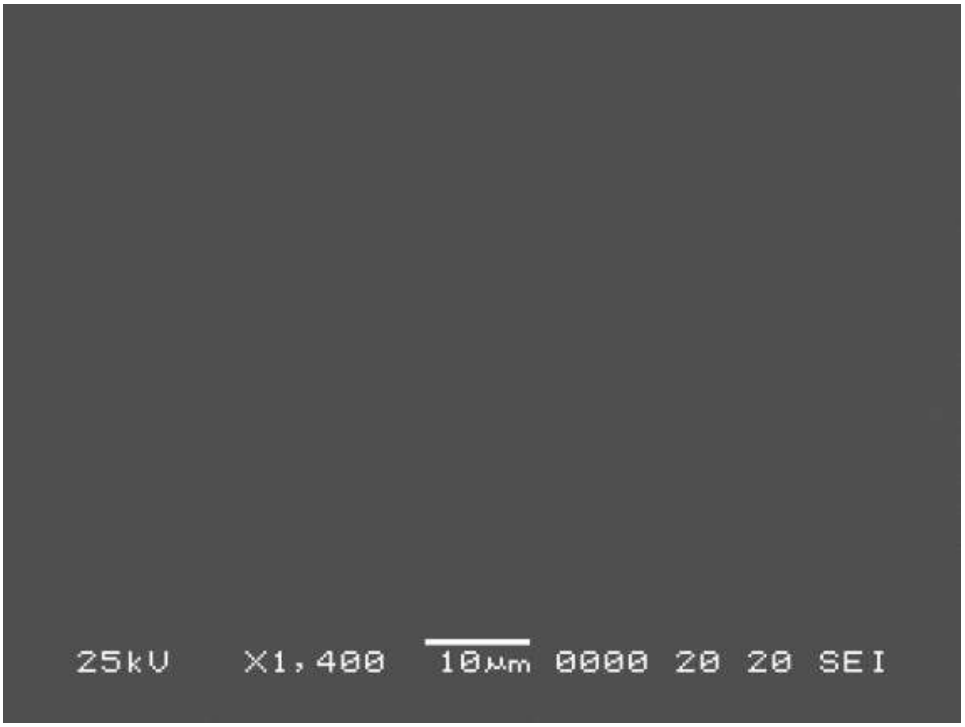

Figure 4 **SEM** micrograph of **Pure Chitosan Film (CS-G00)**

Figure 5 **SEM** micrographs **of** Plasticized Chitosan Film (**CS-G30)**

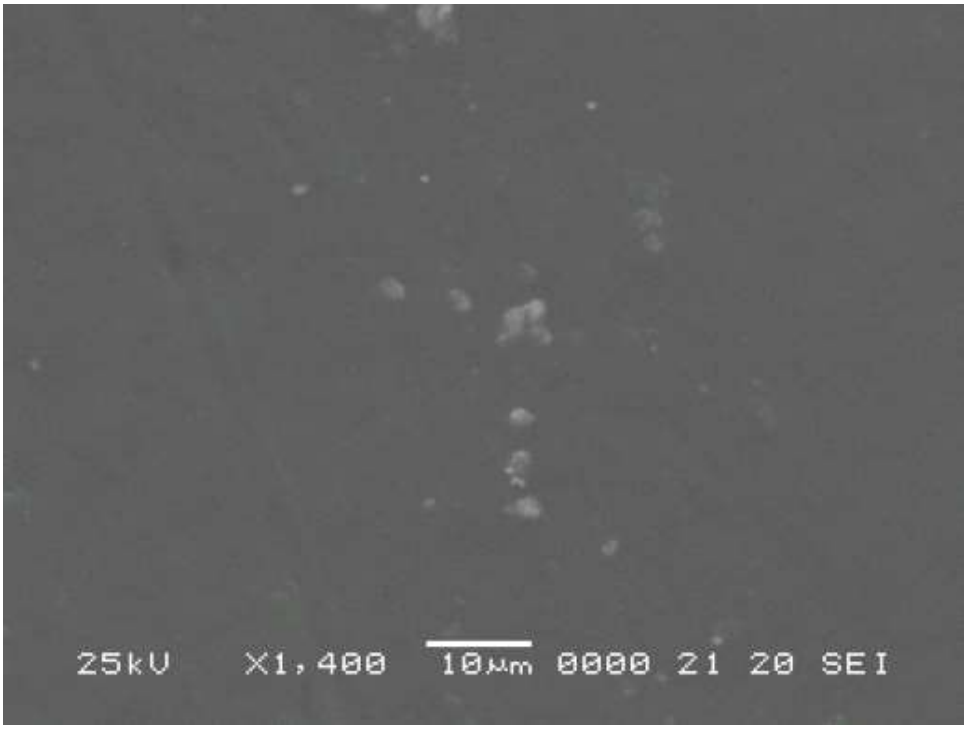

Figure 6 **SEM** micrograph of Plasticized Chitosan Film (**CS-G60)**

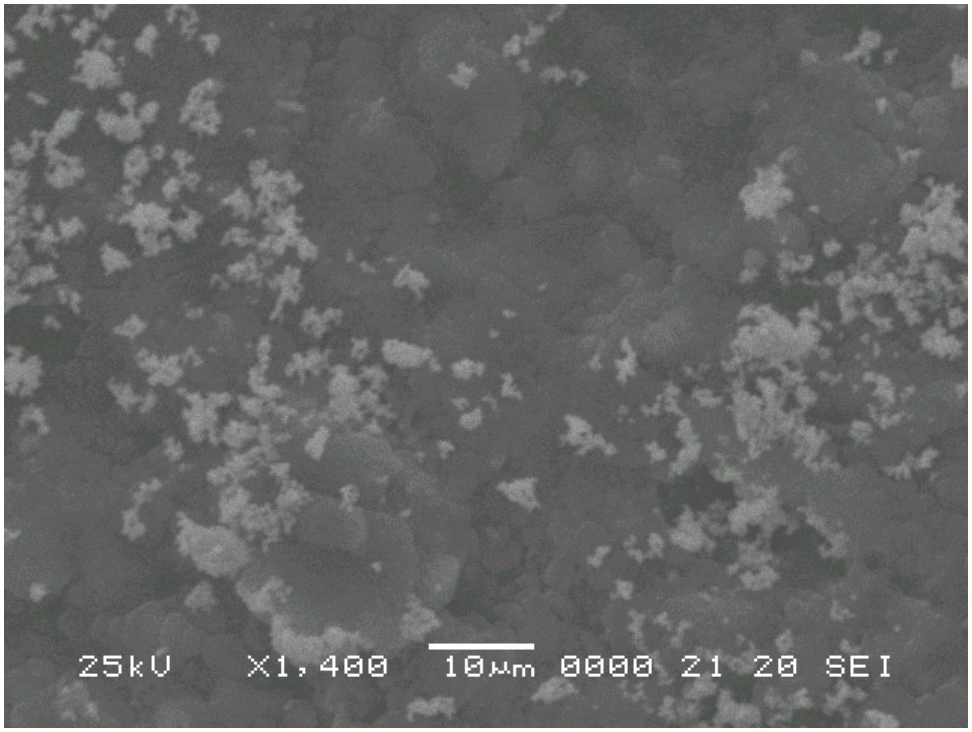

Figure 7 **SEM** micrograph of Plasticized Chitosan Film (**CS-G90)**

3.2 XRD Analysis

As shown in Figure 8 the pure chitosan film CS-G00 found to be amorphous in nature. Wide hump is observed at 2θ=11.23 and 23.92. As a function of doping with different glysrol percentage it become crystallize. Figures 9-11 present the XRD pattern of Chitosan - Glycerol blend under study. For the pure chitosan, there are two peaks around 2θ value 11º and 23°. The maximum crystallization found with 90% glycerol doped samples with 2 θ=11.58, 22.77 and 32.56.

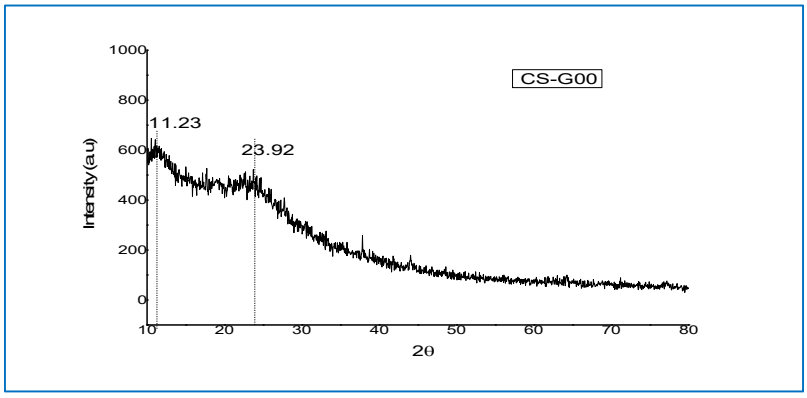

Figure 8 X-ray diffraction pattern of **Pure Chitosan Film (CS-G00)**

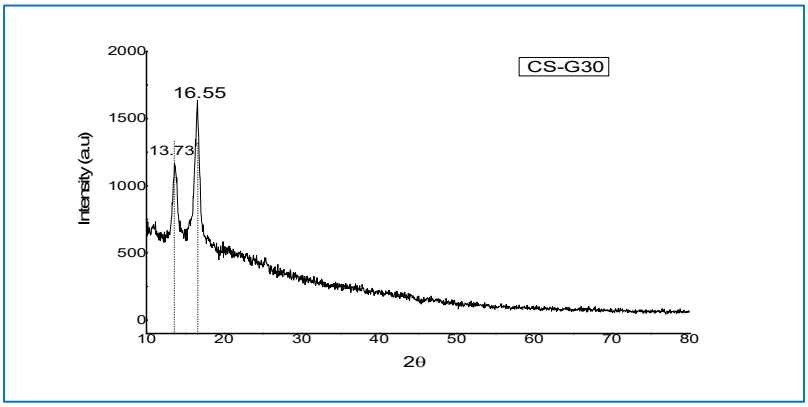

Figure 9 X-ray diffraction pattern of **Plasticized Chitosan Film (CS-G30)**

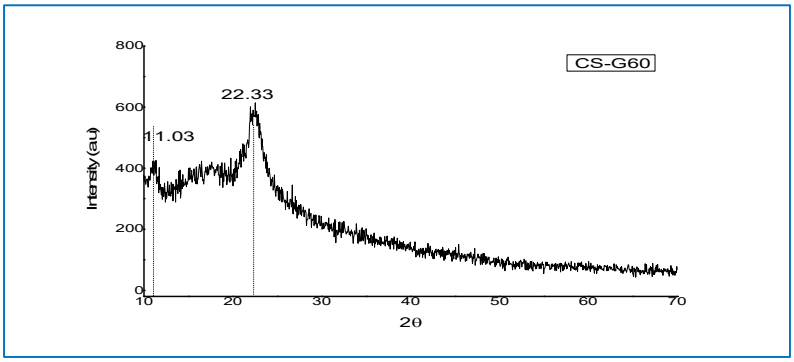

Figure 10 X-ray diffraction pattern of **Plasticized Chitosan Film (CS-G60)**

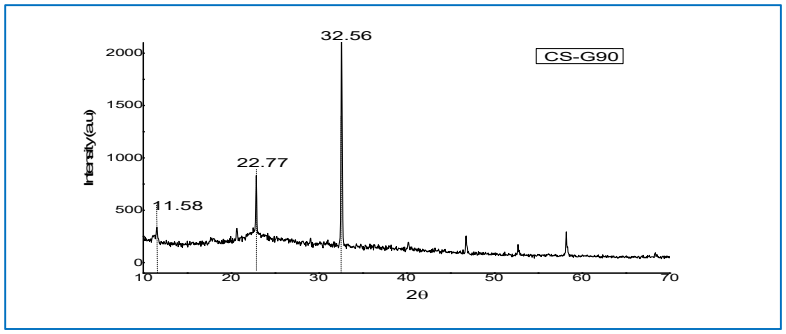

Figure 11 X-ray diffraction pattern of **Plasticized Chitosan Film (CS-G90)**

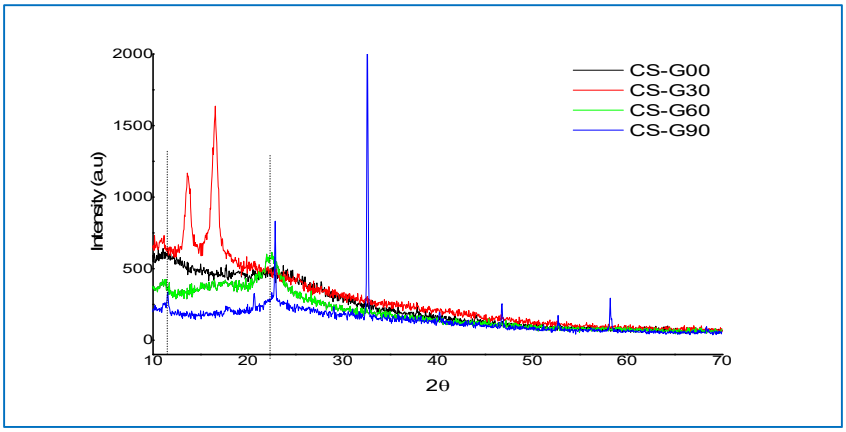

Figure 12 X-ray diffraction pattern of **Chitosan Based Films.**

De Silva et al. 2017[25], presented XRD patterns of chitosan and chitosan/MgO nanocomposite films.The XRD pattern also has one peak about 2θ=23°. The pure chitosan film in the present study shows peak at 2θ=23.92°.Han et al. 2011[19] also reported similar XRD results of pure chitosan film.

4. Conclusions

SEM surface micrographs of Narmada Riverside Crab Shell powder, Chitin powder, Chitosan powder, pure chitosan film and plasticized chitosan films are presented. The SEM photographs of crab shell powder, prepared chitin and chitosan from Narmada riverside crabs exhibited rough and thick surface morphology under electron microscopic examination.. The films produced with the highest proportion of glycerol presented certain structural discontinuities and roughness along the entire surface, as well as small solid particles, contrarily to that present in films with pure chitosan, which had a smooth and very homogeneous surface. It is important to know the material nature whether crystalline or amorphous. X-ray diffraction analysis helps to understand crystallinity and amorphous nature of the materials. Chitosan, like other natural polymers, is known to be partially crystalline polymer where the crystallinity is formed as a result of accumulation of linear chains.

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