

Biodegradation of Paper Wastes by Freshwater Snails: Implications for Management

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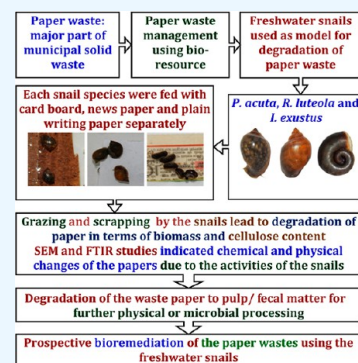
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ABSTRACT: The different types of paper wastes constitute a major portion of municipal solid waste. The present study was aimed to justify the use of freshwater snails for the biological degradation of the paper waste and subsequent availability of cellulose from fecal matter. Three aquatic snails *Racemina luteola*, *Indoplanorbis exustus*, and *Physella acuta* were used to degrade newsprint, cardboard, and common writing paper as paper waste. The consumption of papers by the snail species was subjected to statistical analysis, and the results of Kruskal-Wallis test indicated a significant variation in the paper-specific consumption pattern ($K = 181.762$, $df = 2$, $p < 0.0001$) and no variation in the species-specific pattern. Among the three types of paper used for the study, cardboard was consumed at the highest rate (4.655 ± 0.401 mg per day), and the per capita consumption remained highest for the snail *I. exustus* (2.253 ± 0.273 mg per day). The microscopic images and SEM micrographs of the consumed paper revealed prominent grazing and scraping marks by the snails. The disintegration and relaxation of the papers after consumption indicated that they were on the pathway to degradation. The traditional way of degrading paper waste includes physical, chemical, or microbial treatment of paper pulp. This study represents a novel approach considering the freshwater snails as model organisms for the biodegradation process. Considerable amount of cellulose was present in the fecal samples, which can be extracted and purified for utilization as potent raw material.



INTRODUCTION

The use of nonbiodegradable plastics raises concern about environmental problems. As a substitute/alternative, the papers and cardboards are considered in varied purposes including packaging, bearing the advantage of biodegradability.¹ Owing to the overwhelming use of papers in industries, markets, and households in varied forms such as cardboards, writing paper, newspaper, paper towel, baking paper, and so forth, a huge amount of paper waste is generated in different cities and rural areas. Paper wastes constitute a significant portion of the municipal solid wastes.^{2–7} In India and in many other developing nations, paper wastes are recyclable components in municipal solid waste,³ originating from various sources, with about 34% attributable to the packaging industry.^{5,6} The continued demand and utility of paper materials is evident from the increase of paper waste in municipal solid waste from 3.63% in 1996 to 9.63% in 2011.³ A further increase in paper waste is expected considering the multiple advantages of paper in packaging or other industries.⁶

The paper wastes generated from various sources can be reused as an alternative source of cellulosic raw materials due to their easy availability.^{8,9} Using an appropriate treatment procedure, the paper waste can lead to recycling and reuse in the form of paper pulp to generate several biocommodities. Among the various forms, the paper wastes can be processed physically, chemically, or biologically to extract nano cellulose crystals and fibers.^{8,9} In one sense, the reuse of the paper

wastes will provide an extended lifeline and elongate the life cycle of a product of biological origin. As a waste, the degradation of paper or pulp would have added hazardous material in the environment.

In urban areas, the extensive landfill disposals of the paper wastes and mechanical and physical processed employed for paper degradation may cause potential hazard to the environment and public health.¹⁰ Besides, the paper and pulp mill effluents are also toxic to aquatic ecosystems, affecting the growth, development, reproduction, and behavioral responses of fishes and several other riverine and marine plants and animals.^{11–14} Thus, both manufacture and disposal of papers involve potential environmental hazards, affecting the biota and the environmental quality in general. In order to reduce the possible hazards and sustain the paper production, an effective management of paper wastes is required.^{10,15,16} Among the various modes of thermochemical and mechanical processes of paper and pulp treatment, majority are expensive and unable to degrade specific compounds of paper wastes,

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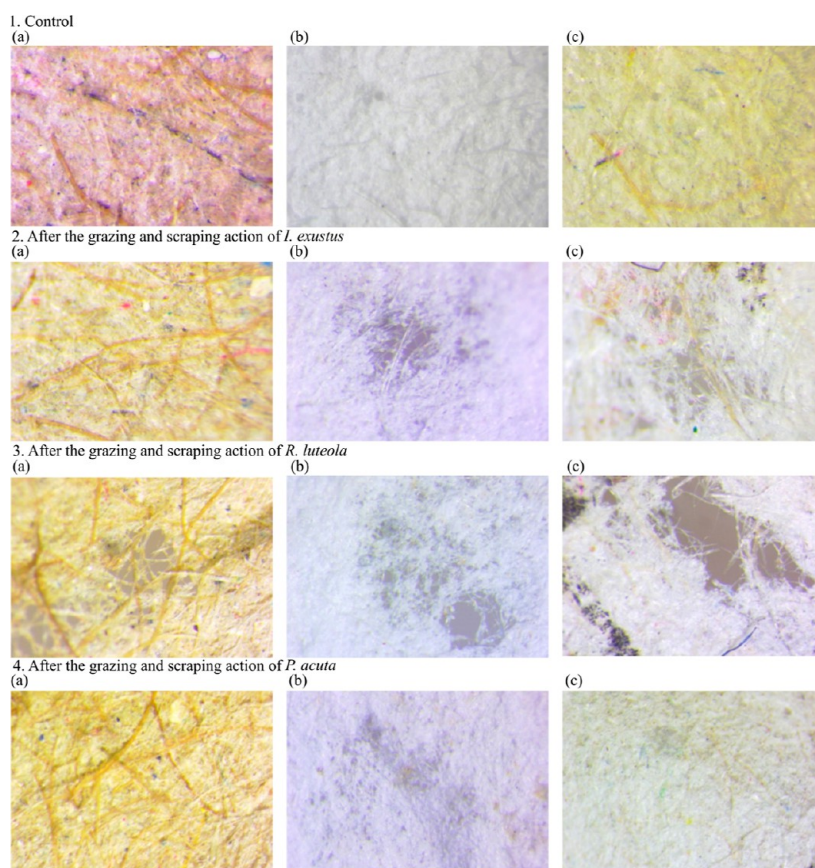


Figure 1. Photographs (magnification 4×) of the surface of paper (a) cardboard, (b) plain paper, and (c) newspaper of (1) control (without the presence of snails) and after 4 days of grazing and scraping activity of (2) *I. exustus*, (3) *R. luteola*, and (4) *P. acuta*.

such as lignin, which persist in a different form following the treatments. In comparison, the application of the biological treatments including vermicomposting is safe environmentally and comparatively cost effective.^{17–19} Such biodegradation, a biologically catalyzed transformation of complex organic compounds into simple molecules,²⁰ facilitates the restoration of the environmental condition with the reduction of environmental pollution.²¹ Owing to the availability of a wide range of biological resources, an advantage of choosing one or many of the biological resources from natural habitat makes it a more feasible process.

In recent times, the use of the cellulase enzyme obtained from the terrestrial snail *Cornu aspersum* (O. F. Müller, 1774) (Gastropoda: Helicidae) in the degradation of the cellulose of the paper wastes into glucose monomers has been reported.²² In view of the significance of recycling of paper wastes, biological degradation seems to be a feasible alternative. The snails and mussels in aquatic systems have long been known as ecological indicators of water pollution.²³ The aquatic snails can monitor water quality by absorbing hazardous metallic substances in their shells and tissue,^{24–26} thus acting as ecosystem engineers.²⁷ In aquatic systems, snails thrive and graze over several substances which float over the water. They are also found to remain attached and thrive on submerged and floating paper materials. Based on this observation, the present study was designed to assess the use of the freshwater snails thriving in the lakes and ponds of West Bengal, India, as a model organism for the degradation of paper waste. In the present study, three freshwater snails, *Racemina luteola* (Lamarck, 1822) (Gastropoda: Lymnaeidae), *Indoplanorbis*

exustus (Deshayes, 1833) (Gastropoda: Planorbidae), and *Physella acuta* (Draparnaud, 1805) (Gastropoda: Physidae) were considered as the model organism for the degradation of the paper wastes. The native snail species *R. luteola* and *I. exustus* are responsible for spreading trematode-borne infections in animals and men.^{28–30} Another snail species, *P. acuta* is an indigenous species of North America but invaded globally in freshwater ecosystems.³¹ These freshwater snails regulate the growth of periphyton, and macrophytes and shells act as the site for algal and periphyton growth.³² The ability of these snail species to degrade the papers of three types, namely, cardboard (CB), newspaper (NP), and plain paper (PP), was assessed along with the amount of the cellulose present in the degraded papers and fecal pellet of the snails fed with several types of paper were also estimated. In general, the degradation of paper waste includes mechanical and physical processes^{16–18} or microbial activity.³³ The utilization of freshwater snails for the biodegradation of paper waste is a pioneer effort. The results are expected to highlight a prospective mechanism of degradation of the paper wastes for either entry as an elemental form in the biogeochemical cycle or enzymatic degradation into sugars. The efficacy of the snails in facilitating the process of paper waste degradation can be judged such that freshwater snails can be considered for the biodegradation process.

RESULTS

All the three species of freshwater snails, namely, *R. luteola*, *I. exustus*, and *P. acuta*, consumed the three different types of paper waste with considerable variations in amount consumed. As a result of the grazing and scraping by the snails during the

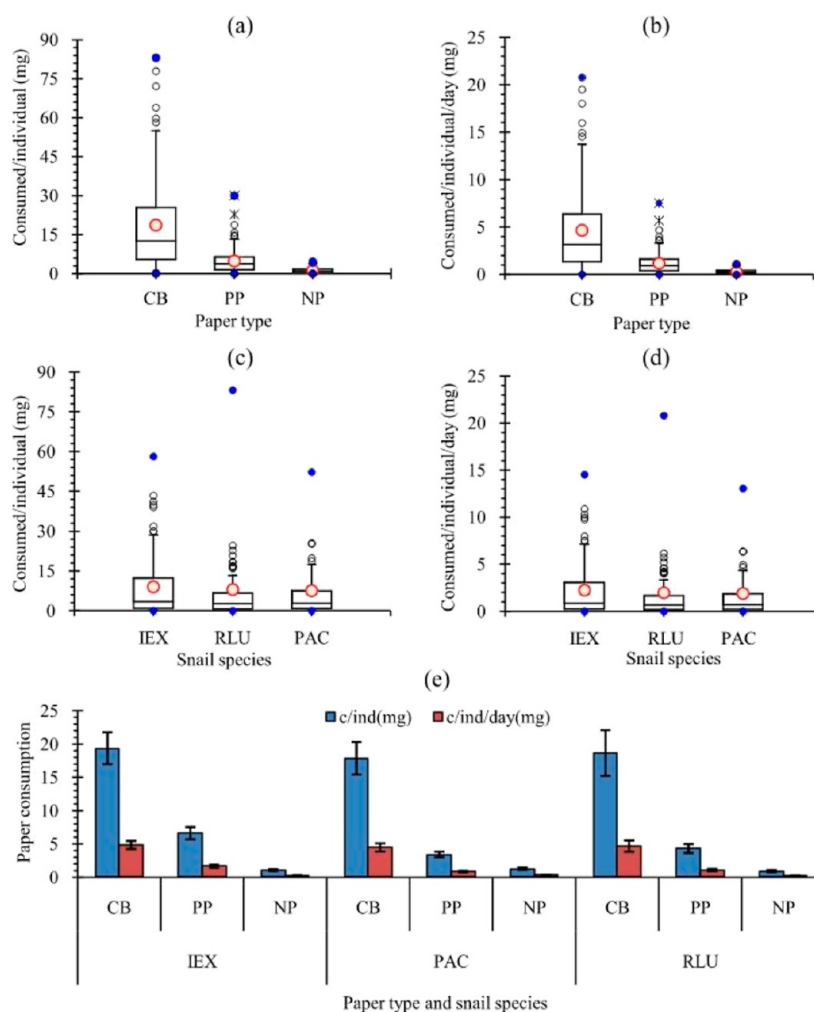


Figure 2. Amount of papers consumed by the snails of similar biomass, expressed in terms of the individuals and the individuals/day. (a,b) Consumption of three snails as function of paper type; (c,d) consumption on three types of paper as function of snail species; (e) bar plot representing the consumption (mean \pm SE) of three types of papers by three snail species [Paper types: CB—cardboard, PP—plain paper, and NP—newspaper; snail species: IEX—*I. exustus*; RLU—*R. luteola* and PAC—*P. acuta*]. SE = Standard error.

feeding activity, prominent signs of degradation of the paper samples were observed (Figure 1). On a comparative scale, the per capita consumption of paper wastes remained highest for the snail *I. exustus* (2.25 ± 0.27 mg/day), followed by *R. luteola* (1.99 ± 0.34 mg/day) and *P. acuta* (1.88 ± 0.26 mg/day) (Figure 2). Among the three types of paper used for the study, the CB was consumed (4.65 ± 0.40 mg/individual/day) at the highest rate, while the NP was consumed (0.27 ± 0.02 mg/individual/day) the least amount by the snails. For the PP, the mean amount consumed was 1.19 ± 0.1 mg/individual/day. The consumption of papers by the snails varied significantly in the paper-specific consumption pattern as observed through the Kruskal–Wallis test ($K = 181.762$, $df = 2$, $p < 0.0001$) but no variation was observed in the species-specific pattern ($K = 2.727$, $df = 2$, $p = 0.256$). Following the consumption, the snail species produced a considerable amount of fecal pellets when CB was consumed (1.32 ± 0.1 mg/individual/day) as food (Figure 3) than for the NP (0.45 ± 0.05 mg/individual/day) or PP (0.84 ± 0.08 mg/individual/day). On a comparative scale, the fecal pellets produced by the snails were highest in *I. exustus*, followed by *R. luteola* and *P. acuta*, respectively. The results of the Kruskal–Wallis test indicated that the fecal matter egested by the snails remained significantly different

based on the paper types ($K = 61.673$, $df = 2$, $p < 0.0001$) and the snail species ($K = 15.982$, $df = 2$, $p = 0.0003$) concerned, but no significant variations were observed in the amount of cellulose in the fecal pellets (Figure 4).

Apparently, following the consumption of paper materials, the snails assimilated some amount of cellulose within the body and released some amount in the fecal matter. The relative mortality of the snails in course of the experiment remained consistent but negligible. The snails produced considerable egg clutches (Figure 5) during the experimental period, indicating that the snails were adapted to the consumption and assimilation of the paper as a source of food. The egg production was high on PP diet for *P. acuta* (20 ± 5.43) and *R. luteola* (46.14 ± 18.40), but *I. exustus* did not oviposit when fed with PP. The fecundity was also high on CB diet with the production of egg clutches 29.7 ± 0.68 by *P. acuta*, 33.36 ± 7.45 by *R. luteola*, and *I. exustus* deposited only one clutch with 17 eggs. The total egg production as a function of snail species and paper types could be presented as follows: total egg = $1 / \{1 + \exp[-(-5.149 - 0.324 * \text{snail species} + 0.253 * \text{paper type})]\}$, with model parameters being significant (intercept = -5.149 ± 0.077 , Wald $\chi^2 = 4428.153$, $p < 0.0001$; snail species = -0.324 ± 0.026 , Wald $\chi^2 = 148.466$, $p < 0.0001$;

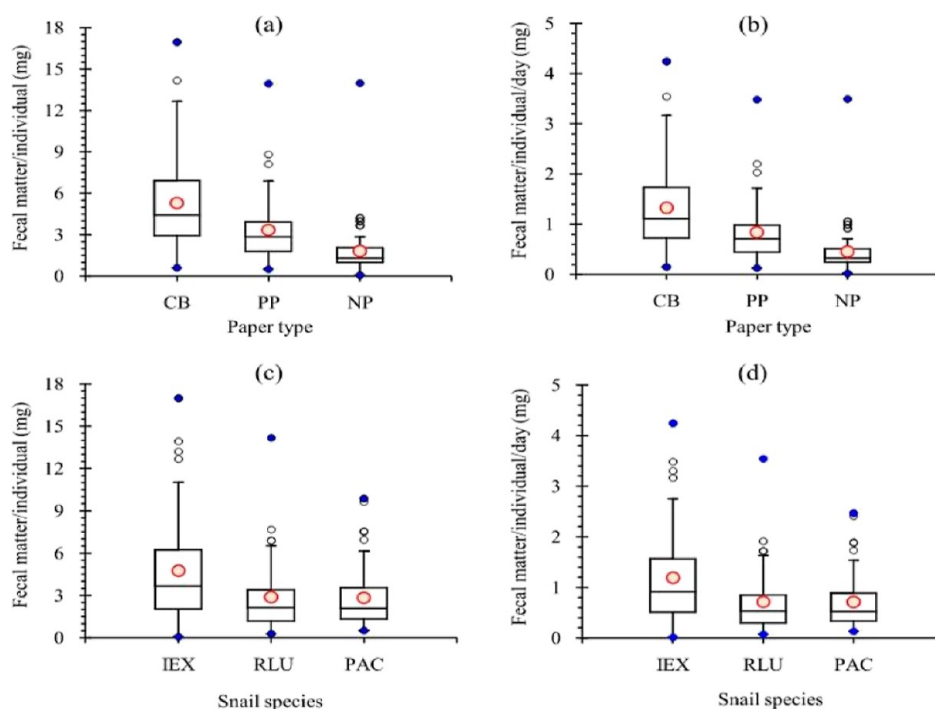


Figure 3. Variations in the fecal matter egested by the snails following the consumption of papers, expressed in terms of the individuals (a,c) and the individuals/day (b,d). [Paper types: CB—cardboard, PP—plain paper, and NP—newspaper; snail species: IEX—*I. exustus*; RLU—*R. luteola* and PAC—*P. acuta*]. The box plot represents the maximum and minimum and mean value (red lined o) of consumption value along with the outliers.

paper type = 0.253 ± 0.026 , Wald $\chi^2 = 92.461$, $p < 0.0001$). The relation of egg per clutch with the snail species and paper types could be presented as follows: $\text{egg/clutch} = 1/\{1 + \exp[-(-6.022 - 0.104 * \text{snail species} + 0.462 * \text{paper type})]\}$, and the model parameters were significant (intercept = -6.022 ± 0.109 , Wald $\chi^2 = 3014.529$, $p < 0.0001$; snail species = -0.104 ± 0.034 , Wald $\chi^2 = 9.29$, $p = 0.002$; paper type = 0.462 ± 0.036 , Wald $\chi^2 = 164.123$, $p < 0.0001$). The regression equation of egg per individual could be presented as follows: $\text{egg/individual} = 1/\{1 + \exp[-(-5.145 - 0.324 * \text{snail species} + 0.253 * \text{paper type})]\}$ with the significant model parameters (intercept = -5.145 ± 0.204 , Wald $\chi^2 = 634.108$, $p < 0.0001$; snail species = -0.324 ± 0.07 , Wald $\chi^2 = 21.209$, $p < 0.0001$; paper type = 0.253 ± 0.069 , Wald $\chi^2 = 13.208$, $p = 0.0002$). Thus, following the consumption of the paper as a food, the snails were able to survive and reproduce and the degradation of the paper was also achieved.

The disintegration and stretching of the fibrous structure of the papers due to the scraping by the snails were prominent in the scanning electron micrographs of the paper samples (Figure 6). The scanning electron micrographs (Figure 7) revealed the presence of paper fibers in the fecal samples also. Figure 8 shows the band intensity and absorbance of CB, PP, and NPs without (control) and after the grazing and scraping of snails, along with the fecal matter after the consumption of respective papers. The CB, PP, and NPs showed the presence of characteristic bands of pyran vibrations at $\sim 800 \text{ cm}^{-1}$, C–H out-of-plane bend at $873\text{--}876 \text{ cm}^{-1}$, aromatic C–H at $1031\text{--}1034 \text{ cm}^{-1}$, C–O–C asymmetric vibrations at $\sim 1162 \text{ cm}^{-1}$, C=O stretch at $\sim 1261 \text{ cm}^{-1}$, and aromatic skeletal vibrations combined with C–H at $\sim 1428 \text{ cm}^{-1}$.^{34–36} In comparison to the spectra of control, change in the wavenumber and/or absorbance of characteristics bands were observed on the

paper samples, which indicated the chemical and physical changes^{35,36} of the papers following the grazing and scraping activities of the snails. An additional band of CH_2 rocking vibration at $\sim 712 \text{ cm}^{-1}$ in cellulose³⁵ was observed on the fecal matters following the consumption of respective papers.

DISCUSSION

In respect to the use of the biological resources, the degradation of the paper wastes appears to be less costly.³⁷ Microbe-derived cellulose degrading enzymes are often utilized for the degeneration of paper materials.^{17,38} The bacterial and fungal species isolated from the several metropolitan wastes produced considerable amount of glucose from the paper wastes.³³ Waste degradation using earthworm *Eisenia foetida* through vermicomposting process is another potential way of waste management.^{39,40} Another study depicted that the degradation of paper cup waste was accelerated through vermicomposting process with bacteria and earthworm *Eudrillus eugeineae*.^{41,42} In recent years, the use of the cellulase enzyme obtained from the terrestrial snail *C. aspersum*^{22,43,44} in degradation of varied types of paper wastes indicates the significance of the snails⁴⁵ in the bioremediation program of recovery and reuse of paper waste at a larger scale. In parity with the propositions of the use of the snails in the bioremediation of the paper waste, the present study is a pioneer effort to establish the potential of the freshwater snails in the recovery and the reuse of the cellulose present in the paper wastes, which otherwise would have added to the total pollution as a solid waste load.

The considerable decrease of weight of experimental papers used as food material of the snails within the experimental mesocosm indicates their ability to reduce the load of paper wastes from the environment. Among the three snail species,

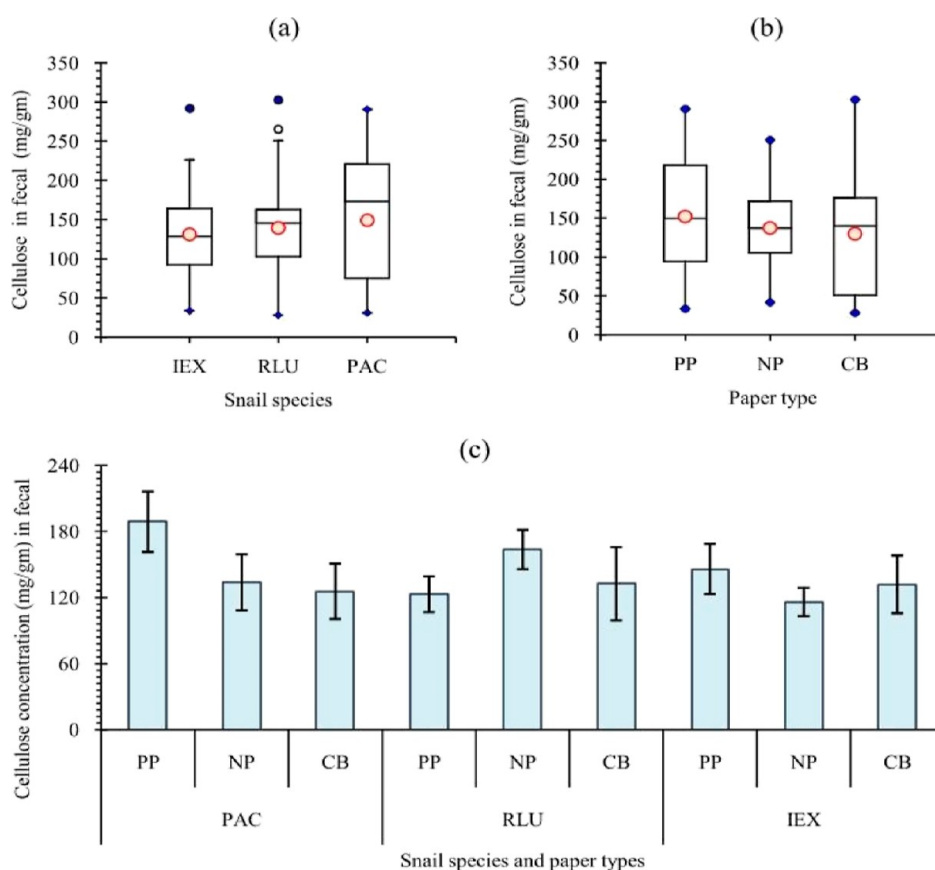


Figure 4. Estimated amount of cellulose content (mean \pm SE) of the fecal matter of the snails fed with different types of papers. (a) Cellulose in the fecal samples as function of snail species; (b) cellulose in the fecal samples as a function of paper types; (c) bar plot representing the consumption (mean \pm SE) of three types of papers by three snail species [paper types: CB—cardboard, PP—plain paper, and NP—newspaper; snail species: IEX—*I. exustus*; RLU—*R. luteola* and PAC—*P. acuta*].

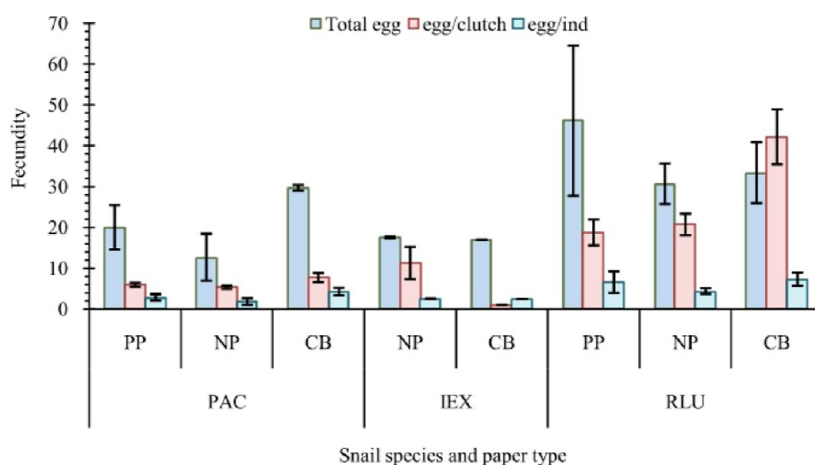


Figure 5. Fecundity of the three snail species (IEX—*I. exustus*; RLU—*R. luteola*; and PAC—*P. acuta*) fed with three different types of papers [CB—cardboard, PP—plain paper, and NP—newspaper].

the consumption rate was highest for *I. exustus*. The CB paper was consumed at a higher rate than other two papers used for the experiment. As shown in the scanning electron microscopy (SEM) pictures, the disintegration and laxation of the paper's fibers were prominent in the consumed paper samples in comparison with the control (kept in water only) paper samples. The cellulosic components of the consumed papers were digested through the enzymatic degradation within the gut of the snails. Part of the cellulosic materials of the

experimental papers were assimilated within the snail body for energy production and the amount of cellulose that had been detected in the fecal matter of the snail species was the undigested part of lignocellulosic materials of the paper. The presence of paper fibers in the fecal pellets of the snails fed with papers was evident from the SEM micrographs. Thus, grazing and scraping by the snails over the paper waste will initiate the primary processing which will augment the degradation process in the natural environment. The release

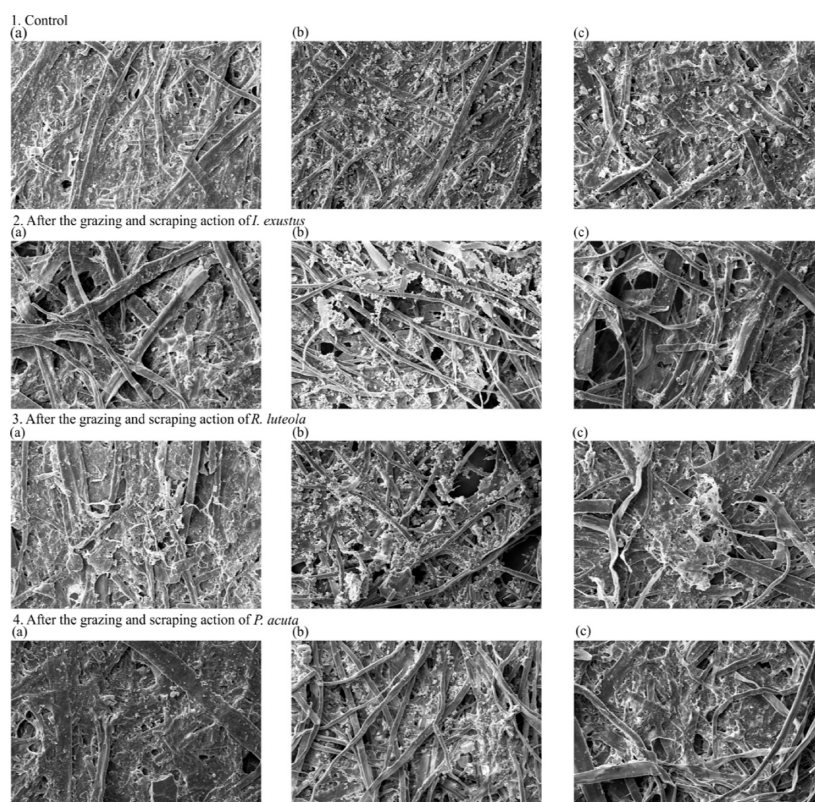


Figure 6. SEM images of the three types of papers (a) cardboard, CB, (b) plain paper, PP, (c) and newspaper, NP of control set (1) and after 4 days of the grazing and scraping activity by *I. exustus* (2), *R. luteola* (3), and *P. acuta* (4). Magnification = 500 \times , Scale bar = 50 μ m.

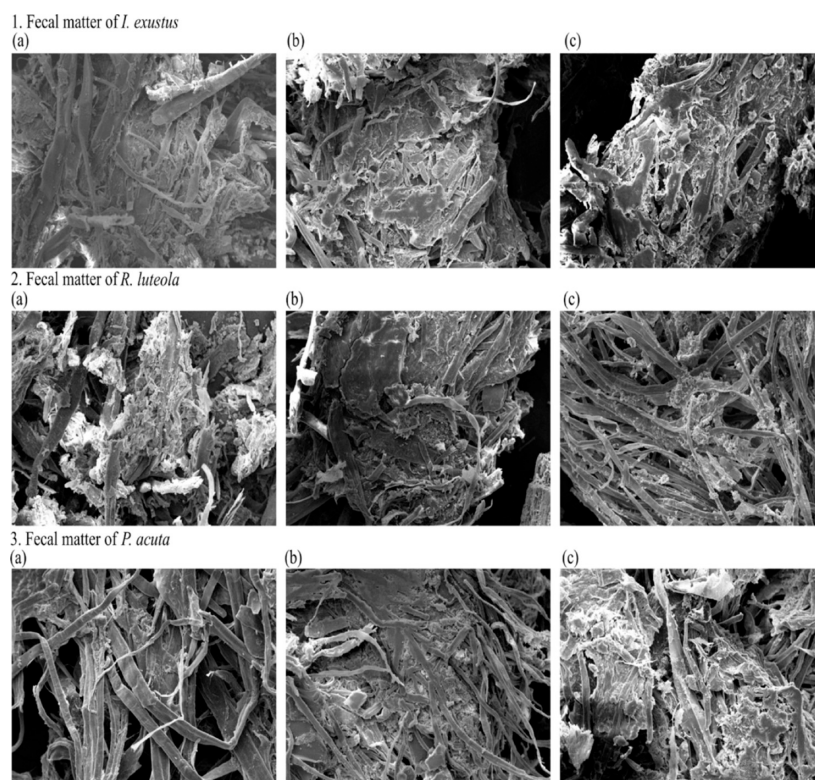


Figure 7. SEM images of the fecal pellets of (1) *I. exustus*, (2) *R. luteola*, and (3) *P. acuta* fed with three types of papers: (a) cardboard - CB, (b) plain paper - PP, and (c) newspaper - NP. Note the looseness of the fibers contrast to those of Figure 6. Magnification = 500 \times , scale bar = 50 μ m.

of loose and disintegrated paper fibers through the fecal pellets will facilitate the microbial growth and enzymatic degradation

or physical transformation. The degradation of paper waste using snails will reduce the biomass and process the solid

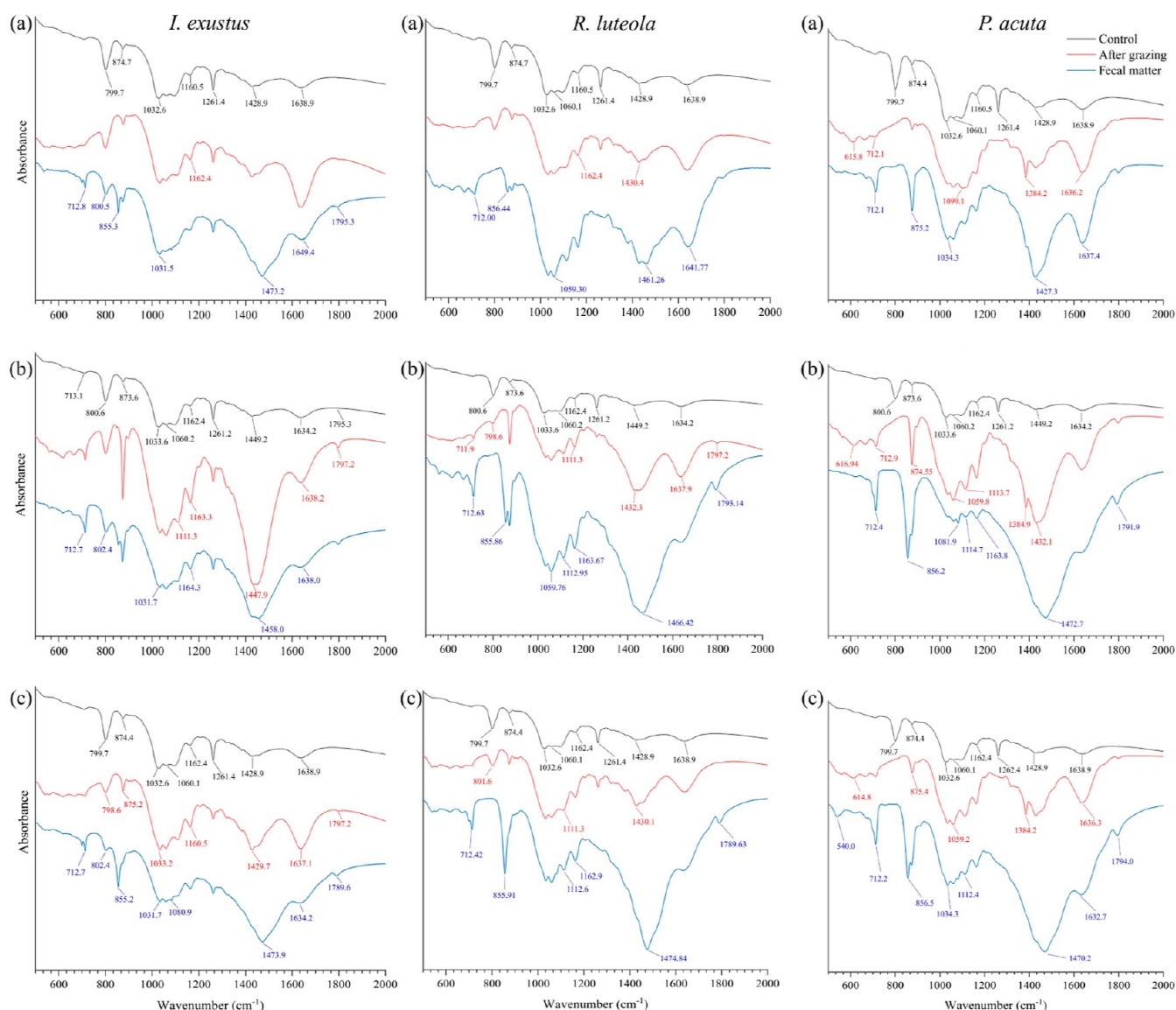


Figure 8. FTIR spectrum of (a) cardboard - CB, (b) plain paper - PP, and (c) newspaper - NPs without (control) and after the grazing and scraping of three snail species, along with the fecal matter after the consumption of respective papers. No absorbance scale is given in the plots as the FTIR spectra of the samples are normalized and shifted parallel to the x -axis.

wastes. The microbial activity on partially processed paper materials will eventually release the cellulose and glucose molecules which will be further degraded by microbes or physical transformation.

Several studies reported that the recycled cellulose fibers from pulp of paper wastes are a promising and sustainable source of biofuel, bio-ethanol,⁴⁶ and other bio-commodities such as porous graphen,⁴⁷ water-soluble fluorescent carbon dots,⁴⁸ graphene oxide quantum dots⁴⁹ multifunctional carbon decorated graphene paper,⁵⁰ porous carbon dots for tetracycline absorption,⁵¹ and metal recover by chemically modified paper waste pulp.^{52,53} The cellulose fibers from paper pulp are also being used as an aerogel to mitigate environmental problems such as oil spill cleaning.^{2,54} The paper wastes are also a promising source of nanocellulose which are now being used for multiple industrial sectors.^{8,9} In the present study, the loose, disintegrated cellulose fibers can be extracted and purified following a proper methodology so that it can be utilized as potent raw material at a commercial level.

The three snail species were able to survive and reproduce consuming only papers as food which indicates their efficacy in degradation of paper wastes. Therefore, further studies may be initiated to judge the efficacy of the snails under varied environmental conditions to degrade and process the paper waste. In addition, future study should be designed to assess the variations of life history traits of snail species fed with papers. Based on the effects of the paper wastes on the life history and ecological traits of freshwater snail species within their aquatic habitats, it can be discerned whether snail species can be effectively used for alleviation of pollutant load in aquatic systems.

CONCLUSIONS

The three freshwater snail species *R. luteola*, *I. exustus*, and *P. acuta* efficiently consumed three types of paper—cardboard, newspaper, and plain writing paper which are common components of municipal solid wastes. The snail species oviposited a considerable number of egg clutches during the

experimental period, reflecting their ability of survival and reproduction while feeding on paper diet only. The amount of cellulose obtained from the fecal pellet of the snails can be extracted and purified for further utilization as raw material in commercial purposes. The degradation of paper waste using snails will reduce the biomass and process the solid wastes. The microbial activity on partially processed paper materials will eventually release the cellulose and glucose molecules which will be further degraded by microbes or by physical transformation.

Though the snail species were able to degrade the paper waste under laboratory conditions, future study should be designed to judge their efficacy in biodegradation of paper waste in natural or semi-natural habitats.

MATERIALS AND METHODS

Snail Collection and Maintenance. The snails were collected from the freshwater habitats in and around Kolkata and adjacent districts of West Bengal, India during the experimental period, between May and September 2018. A nylon insect collection net fitted in a circular iron frame of 40 cm diameter attached with a long handle was dragged along the surface and subsurface (~15–20 cm depth) of the ponds, bogs, and pools for the collection of *R. luteola* and *I. exustus*. The collection of the snail *P. acuta* was made from the sewage drains of the same area using a smaller net fitted to a rectangular frame of 10 × 30 cm size. In all instances, the collections were made from the different sites of the water bodies and the specimens were pooled into a plastic bag and brought to the laboratory. In the laboratory, the snails were placed in a plastic container (Tarson) following segregation of the respective species. The containers were adequately filled with tap water (pH—7.2–7.5) and the snails were maintained at room temperature (27–30 °C). In the course of maintenance of the snails, lettuce leaves were provided *ad libitum* as food. At the end of every 24 h period, the water of the containers was replaced and the dead snails and food remnants were discarded to keep the culture container clean. The collection of the snails was continued during the period of the experiment to replenish the dead snails and keep the population of the respective species at a constant number. In all instances, irrespective of the species, snail individuals were kept in at least a 96 h period in the laboratory for acclimatization prior to their use in the experiments.

Experimental Design. In order to assess the consumption and resultant reduction in the biomass of the papers, three different types of papers were considered, namely, the newspaper (NP, from local newspaper), the plain writing paper (PP, A4 sized paper), and the cardboard (CB, from industrial cartons). The requisite amount of the papers was provided as food to each of the three types of snail species and a long-term assessment was made regarding the change in the biomass and thus degradation. The experiments were carried out in plastic containers of 2 L volume, serving as individual mesocosms. Seven individuals of each snail species were considered for each type of paper. For each snail species and the paper source, six replicates were considered. The size range of the snails used for the experiment was shell length of 8.9–12.1 mm for *R. luteola*, 8–10 mm for *P. acuta*, and 9.1–14.9 mm for *I. exustus*. In a particular mesocosm, the individual snail of a particular species was placed along with a known quantity (representing its dry weight in milligrams) of one type of paper. At the end of the 96 h period, the paper was taken out

and allowed to dry, and subsequently, the biomass was recorded. After that, the paper was placed in the same replicate. The fecal pellets were collected through sieving the water of the mesocosm and allowed to dry for subsequent measurement of the biomass and the analysis of the cellulose content. The observations for all mesocosms were continued for at least seven times for each species and with each type of the paper source. While considering the replicates, the snail individuals and the paper sources were selected following the norms of randomization and interspersions, such that each of the observations constitute a true replicate.⁵⁵

Apart from these six replicates, another set of mesocosms was built with three paper types for each snail species. A control set was considered by placing the only three papers in water. From these mesocosms, after 4 days of consumption, three paper types and fecal matter of the snails were collected and dried in a hot air oven. These consumed paper types along with the papers from the control set were photographed in a binocular microscope (SZ2-ILST, Olympus, Japan) fitted with a digital camera (DGI 510, Dewinter, India).

Cellulose estimation. The amount of the cellulose in the fecal matter was estimated to justify the degradation of the supplied paper. Estimation of the cellulose included acid digestion and removal of the hemicellulose and other components and estimation of the cellulose portion of the concerned material. The method employed for the extraction of lignin, hemicelluloses, and xylosens using acetic acid/nitric acid reagent and the remaining portion, the cellulose is dissolved in sulfuric acid and estimated using the anthrone reagent.^{56,57}

SEM and Fourier Transform Infrared Studies. The CB, PP, and NPs (with and without the grazing and scraping of the snails) along with the fecal matter of the snails were picked from the experimental container on the fourth day. All samples were dried in a hot air oven, and small pieces of papers and few fecal pellets were platinum-coated and respective micrographs were taken by a scanning electron microscope (EVO 18 special edition, Zeiss) to observe the surface structure and morphology. The SEM studies were carried out professionally at CRNN instrument facility, University of Calcutta, Salt Lake campus, Kolkata, India. The completely dried paper samples and fecal pellets (1.0 mg) were mixed with KBr (100 mg, IR spectroscopic grade, Sigma-Aldrich) and thoroughly mixed in mortar and pastels. The 13 mm KBr pellets were made from the mixture using a standard hydraulic press device under a pressure of 100 kN cm⁻² for Fourier transform infrared (FTIR) spectrum analysis. FTIR spectra of the pellets were recorded in the range of 500–2000 cm⁻¹ on a PerkinElmer Spectrum 1000 FTIR Spectrophotometer at 1 cm⁻¹ resolution. A part of the FTIR work was carried out professionally at the Central Instrument Facility, Bose Institute, Kolkata, India.

Data Analysis. The data obtained on the amount of the paper consumed by the snails, the fecal matter produced, and the amount of cellulose in the fecal matter were subjected to non-parametric Kruskal–Wallis test⁵⁸ using the paper and snail types as the explanatory variables. In all instances, the data were represented as per capita consumption or per capita per day consumption and subsequently applied for the statistical analysis using XLSTAT software.⁵⁹ The fecundity of the snails was also recorded during the experimental period in order to assess their ability to reproduce when they were provided with a paper diet. The data obtained on the total eggs, egg per clutch, and egg per individual were subjected to logistic

regression following a binomial generalized linear model with logit link.⁶⁰ The expression of the logistic regression in the form of y (response variable) = $1/\{1 + \exp[-(-a + bx)]\}$ was used to deduce the effect of food type (types of paper) and snail species (explanatory variable) on the fecundity (response variable) of each snail species.

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Conceptualized and compiled by G.A.; chemical analysis and experimentation by G.N.; and data collection by G.N., R.K., A.S., and S.P. Data analysis by G.N. and G.A. FTIR and SEM analysis by P.P. and G.N. Compilation of the article G.N., P.P., and G.A.

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Notes

The authors declare no competing financial interest. The data pertaining to the experiments and results of this article can be made available upon authentic and reasonable request.

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