



Enzymatic scouring of *Kydia calycina* fibers in comparison with chemical scouring

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Abstract: In present study, the *Kydia calycina* fibers were extracted through water retting from young shoots of *Kydia calycina* plant which were collected from G.B.P.U.A. & T., Pantnagar, U.S. Nagar, Uttarakhand. The extracted fibers were scoured with various chemicals (Sodium Carbonate and Sodium Hydroxide) and enzymes (Pectinase, Hemicellulase, Protease and Lipase) and then, the concentration, time, temperature and pH of selected scouring agent were optimized. The one-way ANOVA was used for analyzing the data with help of SAS software. The pectinase enzyme was selected for scouring of fibers on the basis of physical properties i.e. tenacity, elongation and fineness and then scouring variables were optimized as concentration of pectinase enzyme (20%), time (90 minutes), temperature (50°C) and pH (4).

Keywords: Enzymatic scouring, Chemical scouring, Physical property, *Kydia calycina* fibers.

Introduction

Kydia calycina is herbaceous tree of Malvaceae family, which is growing up to 20 m height. The *Kydia calycina* trees, commonly known as *Pula* are distributed in India, Myanmar, Nepal, Pakistan, Bhutan and China. The *Kydia calycina* fibers are one of the natural bast fibers which contain natural impurities along with added impurities on its surface after water retting. These impurities affect directly and indirectly to the properties of fibers, yarn and fabric. So, various scouring agents like chemicals and enzymes are used to remove natural and added impurities from the surface of extracted *Kydia calycina* fibers. Because, the main purpose of scouring is to remove the natural (pectin, lignin, fats and waxes etc.) as well as added impurities (dust or dirt and coloring matters etc.) from the fibers. The removal of these impurities is a pre-processing in order to make good use for end products (Shenai, 1976). According to the Bhattacharya and Shah (2009), scouring of the fibers can be carried out either chemical or microbial. Mojsov, (2012) stated that enzymes are emerging as the best alternative to the polluting textile processing industries because of non-toxic and environmental friendly nature. Enzyme works under mild conditions (pH, temperature) with low water consumption that result in reduced use of harsh chemicals, time, energy and water savings along with improved product quality. Karapinar and Sariisik (2004) reported that various enzymes such as pectinase, protease and cellulase and their combinations were used to remove the impurities in the form of waxes, proteins, pectins, ash and miscellaneous substances such as pigments, hemicelluloses and reducing sugars from cotton. These enzymes and their combinations were used on the basis of 4 to 12 % by weight of present impurities.

Keeping in view of eco-friendly and bio-degradable products, the *Kydia calycina* fibers were extracted from water retting. The extracted *Kydia calycina* fibers were scoured using various scouring agents. The objective of the study was to compare the various scouring agents and to select for best quality *Kydia calycina* fibers.

Materials and Methods

The young shoots of *Kydia calycina* tree were collected from the Agro-forestry Research Center, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar. The various chemicals such as analytical grade sodium hydroxide, sodium carbonate, acetic acid and enzymes like pectinase, hemicellulase, lipase and protease enzymes were used for the study. According to method given by Mauersberger (1954), the fibers were extracted from young shoots of *Kydia calycina* through water retting in 20 days. The temperature and humidity were recorded as $27^{\circ} \pm 2^{\circ}\text{C}$ and $85 \pm 2\%$ respectively. The extracted *Kydia calycina* fibers were scoured with various chemicals, enzymes and their combinations separately. In the chemical scouring, sodium hydroxide and sodium carbonate were used whereas pectinase, hemicellulase, lipase and protease were used in enzymatic scouring. *Kydia calycina* fibers (1 g) were scoured with 2g/l sodium carbonate along with 1g/l non-ionic wetting agent (Lissapol D) at 60°C for 30 minutes. The material liquor ratio was kept as 1:20. After scouring, the fibers were washed and dried (Das, 2009).

In chemical scouring, *Kydia calycina* fibers (1 g) were scoured with 2 % concentration of sodium hydroxide along with 0.5 % concentration of non-ionic wetting agent (Lissapol D) for 30 minutes at boiling temperature. The material liquor ratio was kept 1:30. After that, the fibers were washed and neutralized with 0.2 % concentration of

acetic acid solution for 10 minutes. Again, the fibers were washed under running water and dried (Chattopadhyay *et al.*, 2002).

An enzymatic scouring method was formulated as per available reviews given by different authors. In this method, the impurities such as pectin, protein, lignin, hemicelluloses, fat and waxes were determined in the fibers according to given procedure (Pan, 1999; Alam *et al.*, 2007; Sharma and Goel, 2012). Based on the determined impurities, four different enzymes i.e. pectinase, hemicellulase, lipase and protease and their combinations were used for scouring of extracted fibers separately. The extracted fibers were scoured with 50% concentration of enzymes on the basis of fiber impurities along with 1g/l non-ionic wetting agent (Lissapol D). The temperature of bath was kept at 50° C for 60 minutes. The enzymatic scouring was done in 0.05 M sodium acetate buffer solution of 4.8 pH for pectinase and hemicellulase and same molarity of sodium borate buffer solution of 8.0 pH for protease and lipase enzymes were used. The material liquor ratio was kept as 1:30. The scoured fibers were boiled separately for 10 minutes to inactivate the enzymatic activity. After that, the fibers were washed separately (Diller, 1998; Diller, 1999; Csiszar *et al.*, 2001; kang, 2009).

The physical properties *i.e.* tenacity, elongation, fineness, fiber length and weight loss were determined. The tenacity, elongation and fineness of the *Kydia calycina* fibers were tested using Fafegraph-M. Length of the *Kydia calycina* fibers was analyzed by the single fiber measurement method given by Booth (1996). The weight loss was determined using method given by Garg *et al.* (2013). Data obtained from the experiments were analyzed statistically by using SAS (statistical analysis system) software. Analysis of variance (ANOVA) of group means was computed each of these dependent variables. The significance of means was tested by using Duncun post hoc test at 5 % level of significance. The paired t-test was used to compare the extracted and scoured *Kydia calycina* fibers. The higher the *p*-value means lower the significance. So, we can say that *pd* 0.05 means that the difference is significant.

Results and Discussion

Scouring of *Kydia calycina* fibers: The tenacity, elongation, fineness, fiber length and weight loss of scoured *Kydia calycina* fibers were determined, which are shown in table-1. It is evident from the table-1 that the significant difference was found among

each property of all scoured fibers. The fibers scoured with pectinase (scouring agent 3) had maximum tenacity (4.40 ± 0.46 g/d), elongation (3.43 ± 0.22 %) and length (8.60 ± 0.34 cm) as compared to other scoured fibers (scouring agent 1, 2, 4, 5, 6, 7, 8 and 9). In case of fineness, the denier of pectinase scoured fibers was 26.13±0.44 which is second highest in position among all scoured fibers whereas the weight loss (15.64 ± 0.26 %) of pectinase scoured fiber was found at third position. Therefore, the pectinase was selected as a scouring agent for *Kydia calycina* fibers on the basis of tenacity, elongation and length.

Optimization of scouring variables: A series of experiments were carried out to optimize the variables including concentration, time, temperature and pH for scouring of *Kydia calycina* fibers.

Concentration of pectinase enzyme: The extracted *Kydia calycina* fibers were scoured with various concentrations *i.e.* 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90% and 100% of pectinase enzyme (based on pectin content the fibers) and tenacity, elongation and fineness were tested (Table-2).

The table-2 explains that the significant difference was found among each property of all samples of pectinase scoured *Kydia calycina* fibers. The *Kydia calycina* fibers scoured from 30% concentration of pectinase showed maximum tenacity (4.92 ± 0.07) followed by fiber samples scoured with 20%, 10%, 40%, 50%, 70%, 60%, 80%, 90% and 100%. It was observed that the tenacity of the *Kydia calycina* fibers was increased with the increase in concentration of pectinase up to 30%. After that, the tenacity of the fiber was gradually decreased with increasing concentration of pectinase. The non-significant difference was found among the tenacity of *Kydia calycina* fibers scoured from 10%, 20%, 30%, 40%, 50%, 60% and 70% pectinase enzyme. Therefore, 20 % concentration of pectinase was selected for scouring. The elongation of the *Kydia calycina* fibers scoured with 20% concentration of pectinase was observed highest among all samples. The elongation of this sample was significantly different from other fiber samples scoured with 10%, 30%, 40%, 50%, 60%, 70%, 80%, 90% and 100%. In case of fineness, *Kydia calycina* fibers scoured with 20% concentration of pectinase showed finer than fibers scoured with 10% pectinase and was found coarser than those fiber samples which were scoured from 10%, 30%, 40%, 50%, 60%, 70%,

able-1: Physical properties of *Kydia calycina* fibers scoured with different scouring agents

Scouring agents	Tenacity(g/d) (Mean ± S.E.)	Elongation(%) (Mean ± S.E.)	Fineness(denier) (Mean ± S.E.)	Fiber length (cm) (Mean ± S.E.)	Weight loss (%) (Mean ± S.E.)
Sodium Carbonate	3.84 ^a ± 0.22(18.37)	3.40 ^a ± 0.25(23.19)	31.27 ^{ab} ± 1.71(17.24)	7.51 ^{bcd} ± 0.21(20.25)	18.02 ^a ± 0.24(4.29)
Sodium Hydroxide	4.02 ^a ± 0.23(17.71)	3.06 ^{ab} ± 0.14(14.42)	27.22 ^{bc} ± 1.93(22.38)	7.24 ^{cd} ± 0.17(17.52)	17.95 ^a ± 0.21(3.77)
Pectinase	4.40 ^a ± 0.46(33.23)	3.43 ^a ± 0.22(19.78)	26.13 ^{bc} ± 0.44(5.33)	8.60 ^a ± 0.34(27.91)	15.64 ^a ± 0.26(5.36)
Hemicellulase	3.96 ^a ± 0.18(14.40)	2.85 ^{abc} ± 0.22(24.50)	33.74 ^a ± 1.51(14.13)	7.85 ^{bc} ± 0.13(11.68)	13.73 ^a ± 0.49(11.34)
Protease	3.60 ^{ab} ± 0.25(22.51)	1.88 ^a ± 0.33(54.75)	30.16 ^{abc} ± 2.83(29.70)	7.78 ^{bc} ± 0.17(15.15)	12.44 ^a ± 0.58(14.85)
Lipase	3.63 ^{ab} ± 0.39(33.68)	2.68 ^{bc} ± 0.18(22.35)	33.44 ^a ± 2.38(22.46)	8.13 ^{ab} ± 0.18(16.13)	15.76 ^a ± 0.65(13.09)
Pectinase + Hemicellulase	2.43 ^c ± 0.21(27.74)	1.74 ^a ± 0.07(13.20)	26.49 ^{bc} ± 1.78(21.29)	7.75 ^{bc} ± 0.21(18.86)	17.61 ^a ± 0.37(6.57)
Lipase + Protease	2.90 ^{bc} ± 0.27(29.10)	2.35 ^{bc} ± 0.23(31.45)	26.39 ^{bc} ± 0.87(10.36)	6.97 ^d ± 0.18(18.20)	18.18 ^a ± 0.23(4.00)
Pectinase+Hemicellulase+Lipase+Protease	2.90 ^{bc} ± 0.21(23.07)	2.51 ^{bc} ± 0.16(20.43)	24.74 ^c ± 0.91(11.62)	7.28 ^{cd} ± 0.25(24.66)	17.92 ^a ± 0.28(4.94)
<i>p</i> -values	0.0001	0.0001	0.0009	0.0001	0.0001

Value in parentheses indicate CV (%); Significant difference at 5 % level of significance; If the *p*-value less than 0.05 then significant difference present among mean of all groups within the property; Data followed by same letter within column are not statistically different according to Duncun post hoc test (*p*>0.05)

Table-2: Physical properties of *Kydia calycina* fibers scoured with different concentrations of pectinase enzyme

Concentration of pectinase enzyme (%)	Tenacity (g/d) (Mean ± S.E.)	Elongation (%) (Mean ± S.E.)	Fineness (denier) (Mean ± S.E.)
10	4.63 ^{ab} ± 0.20(13.86)	3.66 ^b ± 0.24(20.80)	31.00 ^a ± 0.55(5.58)
20	4.68 ^{ab} ± 0.17(11.39)	4.54 ^a ± 0.19(13.37)	28.83 ^b ± 0.57(6.25)
30	4.92 ^a ± 0.07(4.32)	3.57 ^{bc} ± 0.32(28.49)	26.52 ^c ± 0.77(9.14)
40	4.61 ^{ab} ± 0.20(14.01)	3.30 ^{bc} ± 0.20(19.21)	25.67 ^c ± 0.63(7.82)
50	4.40 ^{abc} ± 0.46(33.22)	3.43 ^{bc} ± 0.22(19.78)	26.13 ^c ± 0.44(5.33)
60	4.28 ^{abc} ± 0.19(14.35)	2.30 ^{de} ± 0.14(19.36)	25.78 ^c ± 0.95(11.71)
70	4.34 ^{abc} ± 0.22(15.79)	3.06 ^{bc} ± 0.25(26.24)	25.76 ^c ± 0.68(8.37)
80	4.11 ^{bc} ± 0.12(9.38)	3.23 ^{bc} ± 0.20(19.58)	25.92 ^c ± 0.45(5.46)
90	4.06 ^{bc} ± 0.13(9.75)	1.78 ^e ± 0.13(23.04)	25.79 ^c ± 0.68(8.30)
100	3.88 ^c ± 0.12(9.94)	2.92 ^{cd} ± 0.27(29.11)	25.37 ^c ± 0.40(4.96)
p-values	0.0244	0.0001	0.0001

Table-3: Physical properties of *Kydia calycina* fibers pectinase scoured at different time durations

Scouring time (minutes)	Tenacity (g/d) (Mean ± S.E.)	Elongation(%) (Mean ± S.E.)	Fineness (denier) (Mean ± S.E.)
30	3.67 ^{bc} ± 0.13(10.78)	2.90 ^c ± 0.09(10.19)	30.80 ^a ± 0.61(6.28)
60	4.11 ^b ± 0.11(8.47)	3.41 ^b ± 0.11 (10.14)	28.13 ^b ± 0.56(6.33)
90	4.54 ^a ± 0.19(13.16)	4.00 ^a ± 0.20(16.14)	26.36 ^c ± 0.70(8.40)
120	4.07 ^b ± 0.17(13.16)	3.23 ^{bc} ± 0.14(13.43)	25.67 ^c ± 0.50(6.11)
150	3.58 ^c ± 0.13(11.41)	2.95 ^c ± 0.08(8.83)	25.87 ^c ± 0.42(5.16)
p-values	0.0002	0.0001	0.0001

Table-4: Physical properties of *Kydia calycina* fibers scoured at various temperatures

Scouring temp. (Degree Celsius)	Tenacity (g/d) (Mean ± S.E.)	Elongation(%) (Mean ± S.E.)	Fineness (denier) (Mean ± S.E.)
30 ^o	3.56 ^{bc} ± 0.16(13.94)	2.40 ^b ± 0.12(16.01)	24.82 ^b ± 0.33(4.15)
40 ^o	3.94 ^a ± 0.09(7.01)	2.48 ^b ± 0.13(16.23)	25.53 ^{ab} ± 0.36(4.43)
50 ^o	4.43 ^a ± 0.20(14.02)	3.49 ^a ± 0.15(13.80)	26.04 ^{ab} ± 0.50(6.01)
60 ^o	3.81 ^b ± 0.09(7.12)	2.09 ^{bc} ± 0.12(17.75)	26.86 ^a ± 0.21(2.50)
70 ^o	3.37 ^c ± 0.14(13.50)	1.84 ^c ± 0.14(23.77)	24.90 ^b ± 0.76(9.65)
p-values	0.0001	0.0001	0.0191

Table-5: Physical properties of *Kydia calycina* fibers scoured at various pH

Scouring pH	Tenacity (g/d) (Mean ± S.E.)	Elongation (%) (Mean ± S.E.)	Fineness (denier) (Mean ± S.E.)
4	4.90 ^a ± 0.09(5.77)	4.07 ^a ± 0.21(16.08)	25.82 ^{ab} ± 0.50(6.14)
4.4	4.51 ^b ± 0.13(9.14)	3.72 ^{ab} ± 0.23(19.42)	26.86 ^a ± 0.35(4.15)
4.8	4.32 ^b ± 0.20(14.56)	3.41 ^b ± 0.11(10.54)	25.33 ^b ± 0.41(5.12)
5.2	4.16 ^{bc} ± 0.13(9.80)	2.57 ^c ± 0.17(20.53)	26.86 ^a ± 0.21(2.50)
5.6	3.91 ^c ± 0.06(4.94)	2.34 ^c ± 0.12(16.50)	24.90 ^b ± 0.76(9.65)
p-values	0.0001	0.0001	0.0167

Value in parentheses indicate CV (%); Significant difference at 5 % level of significance; If the p-value less than 0.05 then significant difference present among mean of all groups within the property; Data followed by same letter within column are not statistically different according to Duncun post hoc test (p>0.05)

80%, 90% and 100% of pectinase. So, it can be concluded that the 20% concentration of pectinase was selected as optimum concentration on the basis of tenacity and elongation.

Time for scouring: Using optimum concentration of pectinase enzyme, the fiber samples were scoured for 30, 60, 90, 120 and 150 minutes. The physical properties such as tenacity, elongation and fineness of scoured *Kydia calycina* fibers were tested. The results are reported in table-3. The table shows that the

Table-6: Physical properties of extracted and scoured *Kydia calycina* fibers

Physical properties	Extracted fiber	Scoured fiber	p-values
Tenacity (g/d)	4.69	4.90	0.4480
Elongation (%)	4.01	4.07	0.7732
Fineness (denier)	36.34	25.82	0.0001

Significant difference at 5 % level of significance; If the p-value less than 0.05 then significant difference present among mean of all groups within the property

significant differences were found among tenacity, elongation and fineness of fiber scoured for various time i.e. 30, 60, 90, 120 and 150 minutes separately. The *Kydia calycina* fiber scoured for 90 minutes had maximum tenacity (4.54 ± 0.19) and elongation (4.00 ± 0.20) as compared to other fibers which were scoured for 30, 60, 120 and 150 minutes. The *Kydia calycina* fiber scoured for 90 minutes (26.36 ± 0.70) was finer than those which were scoured for 30 and 60 minutes and coarser than the samples scoured for 120 and 150 minutes. Therefore, 90 minutes was selected as optimum time duration for scouring of *Kydia calycina* fiber because of highest tenacity and elongation.

Temperature for scouring: The extracted *Kydia calycina* fibers were scoured with optimum concentration of pectinase enzyme and optimum time of treatment at various temperatures i.e. 30^o, 40^o, 50^o, 60^o and 70^o C and tenacity, elongation and fineness were tested (Table-4). It can be observed from table-4 that significant differences were found among the tenacity, elongation and fineness of fibers scoured at various temperatures. The *Kydia calycina* fiber sample scoured at 50^oC temperature gave maximum tenacity (4.43 ± 0.20) and elongation (3.49 ± 0.15) as compared to other scoured *Kydia calycina* fibers at various temperatures 30^o, 40^o, 60^o and 70^o C. But, the fineness value of *Kydia calycina* fibers scoured at 30^oC had maximum than the fibers scoured at 50^o C temperature. Based on maximum tenacity and elongation, the 50^o C was selected for scouring of extracted *Kydia calycina* fibers.

pH for scouring: After optimizing the concentration of pectinase enzyme, time and temperature of treatment, the *Kydia calycina* fibers were scoured at 4, 4.4, 4.8, 5.2 and 5.6 pH. The tenacity, elongation and fineness of scoured fibers were tested, which are given in table-5. Table-5 shows that there was significant among all physical properties of scoured *Kydia calycina* fibers. The *Kydia calycina* fibers scoured at 4 pH exhibited highest tenacity (4.90 ± 0.09 g/d) than other *Kydia calycina* fibers scoured at 4.4, 4.8, 5.2 and 5.6 pH. In case of fineness, this *Kydia calycina* fibers were coarser (25.82 ± 0.50 denier) as compared to those fibers which were scoured at 4.8 and 5.6 pH but, finer than *Kydia calycina* fibers scoured at 4.4 and 5.2. Therefore, it can be

concluded that the best result can be obtained when scouring was done at 4 pH.

Physical properties of extracted fibers and scoured fibers:

After optimizing the scouring variables i.e. concentration (20%), time (90 min), temperature (50° C) and pH (4), the scoured fibers were compared with extracted *Kydia calycina* fibers on the basis of physical properties, which are given in table-6. Table-6 indicated that the scoured *kydia calycina* fibers had maximum tenacity and elongation and fineness than extracted *kydia calycina* fibers. The result of statistical analysis between extracted and scoured *kydia calycina* fibers showed non-significant difference at five percent level of significant in the tenacity and elongation whereas, significant difference was found in the fineness. But, the physical properties of scoured fibers were found maximum than extracted fibers. Hence, it can be concluded that the scoured *kydia calycina* fibers were better than extracted fiber.

On the basis of physical properties, the scouring with pectinase gave best results among various scouring agents. This may be due to proper removal of impurities from the surface *Kydia calycina* fibers. The pectinase enzyme break down the net structure of pectin and then the pectin content along with other impurities was removed out from fiber surface. This method is substrate specific and it does not alter the cellulose component of *Kydia calycina* fibers. So, the scouring agent i.e. pectinase enhances quality of *Kydia calycina* fibers at optimal condition of scouring variables.

The enzymatic scouring (the scouring with pectinase) of natural unconventional bast fiber i.e. *kydia calycina* fibers becomes more environment-friendly method for processing the *kydia calycina* fibers from young shoot of plant and their disposal is also safe. Therefore, the production of *kydia calycina* fibers can be introduced as an emerging "green" economy.

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