Enzymatic modification of radiata pine kraft fibre and handsheet properties

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SUMMARY

The response of unbleached and bleached radiata pine slabwood and thinnings kraft fibres to treatment with different purified enzymes are quantified and assessed. The enzymes were two xylanases, a cellobiohydrolase, and three endoglucanases.

Unbleached slabwood fibres of high coarseness with thick walls show the greatest response to enzyme treatment. Corresponding fibre and handsheet property differences are smaller for bleached than for unbleached slabwood pulp. Thus, the additional carbohydrate material and/or residual lignin present in the robust fibres of unbleached slabwood pulp must interact in some way during or after enzyme treatment. Some enzyme treatment effects on the unbleached slabwood pulp are –

* Xylanase treated fibres retain their intrinsic strength but can be stiffer with surfaces of lower water affinity and bonding potential compared to untreated fibres. Handsheet properties are modified with tear index being selectively increased.

* Cellobiohydrolase treated fibres increase in size and stiffness but retain their intrinsic strength and bonding potential compared to untreated fibres. Handsheet properties are modified but tear index is not selectively increased.

* Endoglucanase treated fibres (100 µg/g) are degraded with fibre and handsheet strengths lowered. Extents of degradation are different for the three endoglucanases.

Biotechnological processes are becoming increasingly important in the industrial processing of many natural products. Exciting potential exists for biotechnological innovation in the pulp and paper industry as noted and reviewed in the preceding paper of this series (1).

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The understanding of radiata pine fibre properties, how they are affected by processing, and how they relate to end-use requirements is well developed (2,3,4). These understandings are extended in this and the preceding paper (1) through the identification and assessment of the effects of very specific enzymatic treatments on radiata pine kraft fibre properties. Perhaps certain enzymes and/or enzyme treatments can be used to minimize refining requirements, improve pulp drainage properties and/or selectively modify certain paper property relationships. The preceding paper together with that recently published by Pere et al describe the usefulness of a range of analytical techniques for quantifying effects of enzyme treatments on kraft pulp fibres (1,5). In this paper fibre responses to the enzyme treatments are identified, quantified, and interpreted in terms of potential papermaking properties. Unbleached and bleached kraft pulps made from radiata pine thinnings of low fibre coarseness and slabwood of high fibre coarseness are used as substrates for the enzyme treatments.

EXPERIMENTAL

Wood sample origins and kraft pulping

Radiata pine slabwood and thinnings (12 year old) wood samples were from Kaingaroa Forest, New Zealand. The basic density for the slabwood was 460 kg/m³ and thinnings chips 350 kg/m³(1).

Unbleached (Kappa number 27±2) and bleached slabwood and thinnings kraft pulps were prepared using conventional laboratory procedures detailed in the first paper of this series (1).

Enzymes and enzyme treatments

Six purified cellulytic enzymes were obtained from Trichoderma reesei and supplied by Genencor International (Sth San Francisco, CA). The six enzymes were two xylanases labelled E and D, a Cellobiohydrolase (CBH), and three endoglucanases labelled EG-A, EG-B and EG-C. Details of enzyme purity, activity and treatment, and pulp assess-

ment procedures (viscosity, carbohydrate release, and fibre saturation point) are detailed elsewhere (1).

Pulps treated with enzymes were washed and stored at about 20% pulp concentration at 4°C. Residual enzymes remaining in the stored pulps retained some of their activity throughout the period of fibre measurement and pulp evaluation (1).

Handsheet preparation and evaluation

Handsheets were prepared and pulp physical evaluations made in accordance with Appita standard procedures. The load applied during pulp refining with the PFI mill was 3.4 N/mm. Pulps were refined at 10% pulp concentration for 500, 1000, 2000, and 4000 rev. Handsheet physical evaluation data are reported on o.d. bases.

Wet zerospan tensile strengths of untreated and treated pulps were determined according to draft procedure AS/NZS 1301.459rp.

Fibre properties

Microfibril angle was determined using pit apertures as windows according to the procedure of Donaldson (6).

The qualitative presence or absence assessment of the S_1 and S_3 wall layers was determined using combinations of polarized and bright field microscopy. The classification 'S_1 intact' refers to the S_1 layer being intact about the cross-section periphery of a fibre and attached to the S_2 layer. The presence or absence of wall delamination in stained sections was assessed using bright field illumination. A fibre was considered collapsed when fibre thickness was $\leq 0.45 \times$ fibre width. A total of 300 fibres per sample was assessed for each property determination. Fifty fibres on each of six slides were examined for each sample. Slides and pulps were assessed in cyclic order to minimize observer bias. All assessments were made on the same embedded fibres as used in the measurement of fibre cross-section dimensions (7).

Cross-section fibre dimensions of thickness, width, wall area, and wall thickness were measured using image processing procedures (Fig. 1) (7).

Measurements were made on undried fibres, and dried and rewetted fibres from handsheets (AS/NZ 1301.209rp-89). The product of fibre width x fibre thickness represents the minimum fibre cross-section rectangle. The ratio width:thickness is an indicator of the collapse potential of the dried and rewetted fibres. The greater the width and the lower the thickness of a fibre cross-section, the greater is the extent of fibre collapse. Length weighted fibre length and fibre coarseness were determined with a Kajaani FS 200 instrument using TAPPI Method T271 pm-91.

RESULTS AND DISCUSSION

Critical enzyme treatment effects

All the enzyme treatments used caused carbohydrate material to be dissolved compared with the untreated pulps (Table 1) (1). The xylanase treatments are most effective in causing carbohydrate dissolution. The endoglucanase (EG) treatments caused intermediate levels of carbohydrate dissolution and these increased with increasing levels of enzyme treatment concentrations. EG-C is consistently the least effective endoglucanase in causing carbohydrate dissolution. The consistent carbohydrate dissolution brought about by the enzyme treatments is not reflected in decreased fibre coarseness values (Table 2).

Pulp viscosity and wet zerospan tensile index (an indirect measure of intrinsic fibre strength (8)) decrease with increasing endoglucanase concentration but are changed only slightly by the xylanase and cellobiohydrolase treatment (Table 1).

Fibre saturation point trends for the various untreated and treated pulps are unclear (Table 1) (1). With least significant differences between pulps being 0.04 mL/g, however, clear differences exist between the untreated and the treated xylanase-E, CBH and endoglucanase 100 µg/g unbleached slabwood pulps. Differences between the treatments for the bleached slabwood and the thinnings pulp samples are less obvious.

Fibre wall properties and dimensions

Limited quantities of the various enzymes required that only the unbleached slabwood pulp received the full range of enzyme treatments (Table 1).

This slabwood pulp contains fibres which are particularly large in size (width × thickness), and coarse with large wall areas, thick walls and low collapse potential as indicated 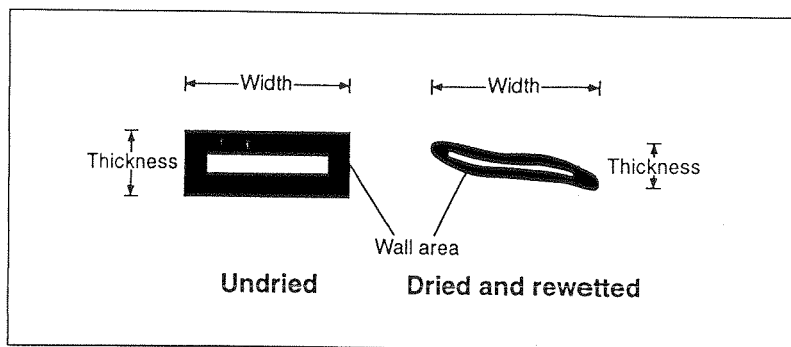by the ratio width:thickness (Table 2). Further-more, the unbleached slabwood pulp shows the greatest response to the enzyme treatments based on changes to fibre cross-section dimensions. The corresponding bleached pulp responds to selected enzyme treatments to a lesser degree, probably because of the absence of lignin and a lower carbohydrate content. The thinnings fibres are short, small in size, and of low coarseness with small wall areas, thin walls, and high collapse potential compared with the slabwood fibres. Fibre dimensions of the bleached and unbleached thinnings are similar.

Fibre wall properties (Table 3) (9)

Fibre wall properties measured include microfibril angle, presence and

Fig. 1　Cross-section diagram of an undried fibre, and a fibre dried and rewetted from a handsheet.

Table 1
Enzyme treatment effects on pulp

Treatment	Solubilized carbohydrate %	Relative viscosity mPa.s	Wet zerospan tensile index[†] N.m/g	Fibre saturation point, mL/g
Unbleached slabwood				
Untreated	0.02	31.2	166	1.44
Xylanase-E, 400 µg/g	1.54	30.3	169	1.35
Xylanase-D, 400 µg/g	1.35	28.6	150	1.40
CBH[*], 400 µg/g	0.11	28.3	160	1.36
EG[*]-A, 400 µg/g	0.46	12.8	54	1.51
EG-B, 400 µg/g	0.74	18.3	81	1.48
EG-C, 400 µg/g	0.42	14.5	42	1.39
EG-A, 100 µg/g	0.16	19.1	89	1.39
EG-B, 100 µg/g	0.33	24.8	114	1.37
EG-C, 100 µg/g	0.14	17.9	72	1.40
Bleached slabwood				
Untreated	0.10	16.3	150	1.36
Xylanase -E, 400 µg/g	2.17	15.8	151	1.33
CBH, 400 µg/g	0.32	15.9	140	1.33
EG-A, 100 µg/g	0.27	10.4	72	1.35
EG-B, 100 µg/g	0.27	14.1	127	1.30
EG-C, 100 µg/g	0.13	12.0	100	1.14
Unbleached thinnings				
Untreated	0.02	22.9	109	1.45
EG-A, 100 µg/g	0.23	12.7	49	1.25
EG-B, 100 µg/g	0.44	17.6	70	1.17
EG-C, 100 µg/g	0.20	12.6	45	1.75
Bleached thinnings				
Untreated	0.08	16.1	106	1.34
EG-A, 100 µg/g	0.26	10.5	49	1.44
EG-B, 100 µg/g	0.34	14.6	93	1.46
EG-C, 100 µg/g	0.15	12.6	68	1.40
LSD[‡]		0.40		0.04

* CBH – Cellobiohydrolase, EG – Endoglucanase
† Wet zerospan tensile index determined at 500 PFI mill revolutions
‡ Least significant difference between means at the 95% level of significance

Table 2
Fibre dimensions of dried and rewetted pulps

Treatment	Length mm†	Coarseness‡	Cross-section dimensions					
			Width	Thickness	Width x thickness	Wall area	Wall thickness	Width: thickness
		mg/m	μm		μm²	μm²	μm²	
Unbleached slabwood								
Untreated	2.82	0.250	30.9	12.2	378	223	3.70	2.68
Xylanase-E, 400 μg/g	2.88	0.265	32.0	11.8	375	228	3.79	2.91
Xylanase-D, 400 μg/g	2.90	0.253	32.2	11.8	382	240	3.99	2.91
CBH*, 400 μg/g	2.90	0.267	32.5	12.6	416	246	3.89	2.75
EG*-A, 400 μg/g	2.77	0.260	32.1	11.3	367	217	3.46	2.99
EG-B, 400 μg/g	2.85	0.253	30.7	11.7	357	211	3.47	2.87
EG-C, 400 μg/g	2.77	0.270	34.3	12.4	425	257	3.83	2.94
EG-A, 100 μg/g	2.85	0.246	35.8	14.0	499	305	4.32	2.75
EG-B, 100 μg/g	2.87	0.259	35.0	12.6	448	263	3.74	2.93
EG-C, 100 μg/g	2.84	0.261	35.3	12.2	432	255	3.63	3.10
Bleached slabwood								
Untreated	2.82	0.257	32.8	11.6	380	229	3.53	3.04
Xylanase-E, 400 μg/g	2.75	0.268	33.0	11.2	371	232	3.71	3.14
CBH, 400 μg/g	2.78	0.257	31.4	11.9	370	222	3.59	2.87
EG-A, 100 μg/g	2.75	0.259	31.5	11.7	364	219	3.57	2.93
EG-B, 100 μg/g	2.78	0.268	33.2	11.8	389	232	3.62	3.07
EG-C, 100 μg/g	2.80	0.257	32.8	11.3	358	211	3.31	3.21
Unbleached thinnings								
Untreated	1.99	0.180	31.8	10.2	327	177	2.86	3.38
EG-A, 100 μg/g	1.93	0.179	32.8	10.4	337	186	2.85	3.47
EG-B, 100 μg/g	1.95	0.181	31.7	10.7	340	185	2.94	3.25
EG-C, 100 μg/g	1.92	0.186	31.0	10.1	315	184	3.01	3.30
Bleached thinnings								
Untreated	1.94	0.178	31.7	10.7	340	193	3.11	3.20
EG-A, 100 μg/g	1.88	0.180	31.8	11.1	350	198	3.19	3.12
EG-B, 100 μg/g	1.91	0.177	33.3	10.5	345	198	3.01	3.52
EG-C, 100 μg/g	1.90	0.184	31.2	10.5	325	183	2.95	3.29
LSD#			1.7	0.7	30	15	0.2	0.2

*   CBH – Cellobiohydrolase, EG – Endoglucanase
†   Length weighted fibre length: Confidence limits 0.05 mm at the 95% level of significance based on the PAPRO Standard Medium calibration pulp. Two length determinations made for each pulp, with 10000–15000 particles measured in each (PAPRO Standard Method 1.306)
‡   Fibre coarseness: Confidence limits 0.015 mg/m at the 95% level of significance based on the PAPRO Standard Medium calibration pulp. Two determinations made for each pulp (PAPRO Standard Method 1.306)
\#   Least significant difference between means at the 95% level of significance

Fig. 2   Unbleached slabwood enzyme treatment, handsheet apparent density and PFI mill refining level relationships (xylanases and CBH, 400 μg/g; EG-A; B and C, 100 μg/g).

condition of the S_1 and S_3 layers, incidence of wall delamination, thick walled or 'latewood' proportions, and collapse degrees. Except for microfibril angle, all properties measured are different for the untreated and enzyme treated pulps but similar for all enzyme treatments, both when the fibres are undried or dried and rewetted from handsheets (Table 3). Microfibril angles for the dried and rewetted, and undried pulps are unchanged by the enzyme treatments.

Dried and rewetted fibres from handsheets (Table 2)

The dimensions of fibres dried and rewetted from handsheets have been modified by the pressing and drying sheetmaking process (Fig. 1) (11,12).

The dried and rewetted unbleached slabwood fibres respond to the various enzyme treatments as follows (Table 2) –

• Fibre length and coarseness are unchanged by the enzyme treatments. The Kajaani FS 200 pulp fibre coarseness measurement procedure is insufficiently sensitive to show differences associated with carbohydrate losses brought about by the enzyme treatments (Table 1).

• The two xylanase treatments cause fibre width to increase and fibre thickness to decrease in accordance with an increased collapse potential. This occurs with minimal increases (not significant for xylanase-E), rather than the expected decreases in wall area and wall thickness as a result of carbohydrate dissolution with xylanase treatment (Table 1).

• CBH treatment causes fibre wall areas to increase, and fibre cross-section areas to increase substantially in the width and thickness dimensions so that fibres remain essentially uncollapsed relative to untreated fibres. Such effects could indicate fibre and fibre wall resistance to collapse and contraction during handsheet pressing and drying processes rather than to extensive fibre swelling resulting from CBH treatment (Tables 2,4).

• The three endoglucanase treatments at 100 μg/g show remarkable effects on dried and rewetted fibres. They are extremely large in size and wall area but generally more collapsed than the untreated fibres. Again such fibres are possibly resistant to contraction during sheetmaking rather than physically swollen by the enzyme treatments themselves (Tables 2,4). Fibres treated
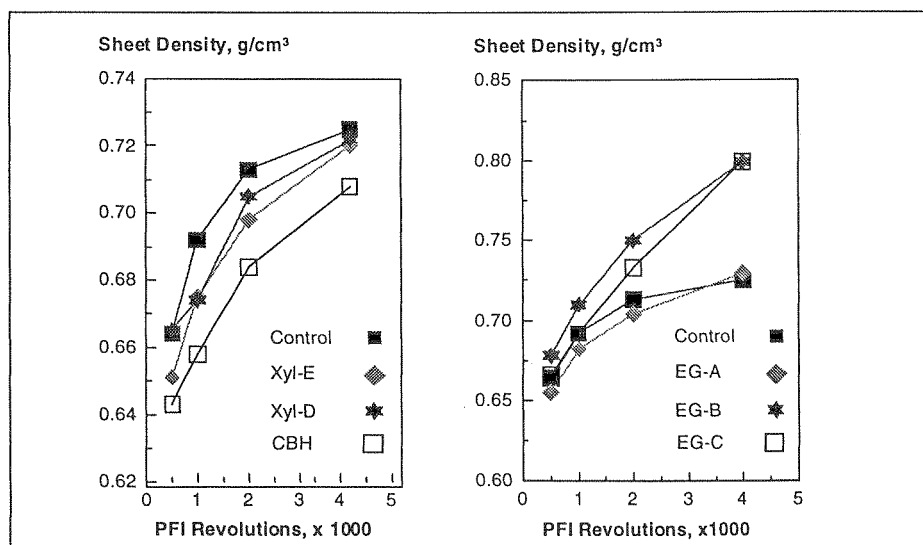
with endoglucanase at 400 µg/g have cross-section dimensions similar to those of the untreated fibres except for the EG-C fibres which show similar dimensions for both enzyme treatment concentrations.

Enzyme treatment effects on the unbleached and bleached slabwood pulps are very different (Table 2). Enzyme treatment effects on the bleached pulp fibre dimensions are generally not significant compared with untreated fibres. The EG-A and EG-C endoglucanase treatments do, however, show marginal decreases in fibre size and wall area reminiscent of the 400 µg/g unbleached fibre treatment effects. Additional carbohydrate material and/or residual lignin in the unbleached slabwood fibres are somehow able to influence their response to enzyme treatments and/or to pressing and drying during sheetmaking.

Fibre dimensions of the bleached and unbleached thinnings pulps are generally similar and unchanged by the endoglucanase treatments. These thinnings fibres are of extremely high collapse potential even without modification using enzymes (Table 2) (10). It is fortunate that the slabwood fibres used represent the high end of the radiata pine fibre coarseness and wall thickness range and show measurable responses to selected enzyme treatments.

Undried fibres (Table 3)

This undried fibre classification refers to fibres which are never dried after kraft pulping, and before and after enzyme treatment. Pulps are, however, stored in a crumbed state at 20–25% solids at 4°C before and after enzyme treatment. Undried fibres are uncollapsed with large cross-section areas, and walls are swollen and porous compared with dried and rewetted fibres (Tables 2,3) (11).

The undried unbleached slabwood fibres respond to the various enzyme treatments as follows (Table 3) –

• The xylanase treatments show minimal effects on undried fibre dimensions: fibre wall area and thickness are unchanged, while fibre thickness is decreased and the related properties of shape and cross-section area correspondingly modified. This decrease in undried fibre thickness is a real effect of xylanase treatment since it is retained in the dried and rewetted fibres with high collapse potentials (Table 2).

Table 3
Fibre dimensions of undried pulps

Treatment*	Fibre cross-section dimensions					
	Width µm	Thickness µm	Width x Thickness µm²	Wall area µm²	Wall thickness µm	Wall: thickness µm
Unbleached slabwood						
Untreated	31.1	17.6	561	276	3.88	1.87
Xylanase-E, 400 µg/g	32.1	16.2	531	284	3.95	2.13
Xylanase-D, 400 µg/g	32.0	16.4	536	274	3.82	2.06
CBH*, 400 µg/g	32.1	16.0	523	269	3.73	2.15
EG*-A, 400 µg/g	30.8	15.9	496	258	3.74	2.11
EG-B, 400 µg/g	32.3	15.9	518	275	3.83	2.21
EG-C, 400 µg/g	32.1	16.3	532	284	4.03	2.12
EG-A, 100 µg/g	32.3	16.3	533	292	4.17	2.13
EG-B, 100 µg/g	32.7	16.5	542	289	4.13	2.13
EG-C, 100 µg/g	32.9	16.4	539	290	4.08	2.17
Bleached slabwood						
Untreated	33.4	14.3	481	258	3.60	2.54
Xylanase - E, 400 µg/g	31.3	13.3	414	236	3.62	2.55
CBH, 400 µg/g	32.0	15.0	481	253	3.62	2.35
EG-A, 100 µg/g	32.0	15.0	487	261	3.71	2.33
EG-B, 100 µg/g	33.0	15.5	521	276	3.75	2.28
EG-C, 100 µg/g	32.6	15.1	499	269	3.73	2.34
Unbleached thinnings						
Untreated	30.3	15.8	482	241	3.59	2.05
EG-A, 100 µg/g	31.3	14.7	471	227	3.24	2.31
EG-B, 100 µg/g	30.5	13.9	426	217	3.29	2.37
EG-C, 100 µg/g	31.1	13.5	426	215	3.18	2.52
Bleached thinnings						
Untreated	31.2	13.5	427	218	3.25	2.51
EG-A, 100 µg/g	29.2	12.7	370	192	3.04	2.59
EG-B, 100 µg/g	29.8	13.8	415	211	3.21	2.38
EG-C, 100 µg/g	31.3	14.2	448	224	3.23	2.37
LSD†	1.7	0.9	42	20	0.2	0.2

* CBH – Cellobiohydrolase, EG – Endoglucanase
† Least significant difference between means at the 95 % level of significance
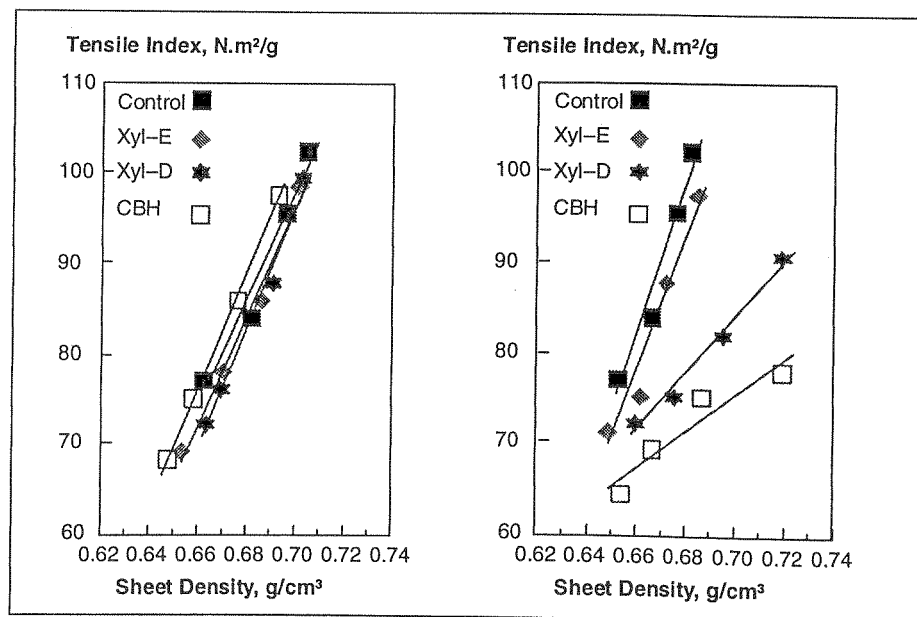
Fig. 3 Unbleached slabwood enzyme treatment, and handsheet tensile index and apparent density relationships (xylanases and CBH, 400 µg/g; EG-A, B and C, 100 µg/g).

- CBH treatment effects are similar to those of the xylanase treatments: fibre thickness and cross-section area are decreased while wall area and thickness are unchanged. Such similar fibre dimensions for the CBH and xylanase treatments are, however, confined to the undried fibres only. CBH treated fibres and fibre walls are able to resist collapse/contraction during sheetmaking pressing and drying processes since dried and rewetted fibres remain uncollapsed and their walls remain swollen (Table 2).

- The endoglucanses all cause fibre thickness and the related cross-section area to decrease. This effect is least for the 100 µg/g and greatest for the 400 µg/g treatments. The three 100 µg/g, and the EG-C 400 µg/g treatments have consistently high wall areas and wall thicknesses compared with all others. It is noteworthy that these four treatments also show the largest dried and rewetted fibres and walls after sheetmaking (Table 2). Hence these fibres are able to resist both fibre and wall contraction during sheetmaking but in a way different to that caused by CBH treatment. The dried and rewetted fibres of the four endoglucanase treatments (3 x 100 µg/g, and EG-C 400 µg/g) are markedly larger in the width dimension compared with the CBH and untreated fibres. Fibre saturation points for the CBH and four endoglucanase treatments are consistently low compared with all others (Table 1).

Undried bleached fibres (slabwood and thinnings) without enzyme treatment are of low thickness, cross-section area, wall area, and wall thickness but of high collapse potential compared with corresponding unbleached fibres (Table 3).

For the bleached slabwood fibres the CBH and endoglucanase treatments show consistent but marginal increases rather than decreases in thickness dimension, with other fibre properties generally not significantly different from those of the untreated pulp (Table 3).

Treatment of the bleached and unbleached thinnings pulps with endoglucanase show differences which generally parallel those of the slabwood pulps, although most are not significant (Table 3). With the endoglucanase treatments, fibre thickness generally decreases for the unbleached pulp, and is unchanged for the bleached pulp.

FIBRE AND HANDSHEET PROPERTY INTERRELATIONSHIPS

Handsheet apparent density

Handsheet apparent density is primarily determined by fibre packing arrangements which in turn are influenced by fibre dimensions, coarseness, and collapse, as well as the bonding potentials of fibre surfaces and their influence on overall web consolidation. All of these fibre properties can be expected in some way or another to be modified by selected enzyme treatments. Hence apparent density is considered the critical handsheet property for enzyme treatment assessment. Other properties such as tensile strength, light scattering coefficient, and tearing resistance are all critically dependent on handsheet apparent density or bulk (3).

Unbleached slabwood: Handsheet apparent density decreases with the xylanase and CBH treatments, is unchanged with the EG-B 100 µg/g treatment, and increases with the EG-A and EG-C 100 µg/g treatments (Fig. 2) (9). Increases in apparent density brought about by pulp refining are roughly linear for the EG-A and EG-C treatments whereas those for the untreated and all other treatments are logarithmic as expected.

Enzyme treatment, fibre property and handsheet apparent density interrelationships are explained as follows –

Xylanase treated fibres retain their relative viscosity and wet zerospan tensile strength (Table 1) but are more collapsed than those in untreated handsheets (Table 2). Also, some selective xylan dissolution occurs relative to the other enzyme treatments (Table 1) (1). Increased fibre collapse could be expected to increase handsheet density. In contrast, the loss of xylan containing carbohydrates could be expected to make fibre surfaces less hydrophilic to increase fibre stiffness and decrease bonding potential, and thus decrease handsheet density. Xylan has been shown to be concentrated in the outermost layers of kraft fibres (12). A lowering of the fibre saturation point from 1.44 to 1.35 mL/g for the xylanase-E treatment (Table 1) suggests a contraction of fibre wall pore volume and a stiffening of fibre walls. The marginal increase in fibre collapse brought about by xylanase treatment is more than counteracted by the influence on packing density and consolidation of increased fibre stiffness and bonding potential.

- CBH treated fibres also retain their relative viscosity and wet zerospan

tensile strength (Table 1) but are resistant to collapse and their fibres and walls remain expanded compared with corresponding untreated dried and rewetted fibres. Hence, the marked decreases in handsheet density are to be expected. It is of interest that the selected response of CBH treatment is evident only from the measurement of fibres rewetted from handsheets. Conventional assessment of undried CBH treated fibres shows them to be similar to all others except for a significant decrease in fibre saturation point (Table 1).

- Fibres treated with endoglucanase at 100 µg/g show minimal increases in handsheet density relative to the untreated fibres when compared at the low 500 rev PFI mill refining level (Fig. 2). At higher levels of pulp refining handsheet densities are increased greatly for EG-A and EG-C, but remain similar to the untreated pulp for EG-B. The low fibre strengths of EG-A and EG-C, indicated by low relative viscosities and zerospan tensile strengths (Table 1), accounts for their ease of refining and high handsheet density values. In contrast corresponding fibre strength indicators for EG-B are substantially higher and handsheet densities are similar to those of the untreated pulp. Fibre dimensions for the three endoglucanase treatments are generally similar (Tables 2, 3) and do not reflect strength and handsheet apparent density differences between EG-B, and EG-A and C. The exceptionally large width dimensions of the dried and rewetted endoglucanase treated fibres (Table 2) suggest that the integrity of the fibre has been lost through extensive disruption/weakening of cellulose wall structures.

- Fibres treated with endoglucanase at 400 µg/g are extensively damaged as indicated by their relative viscosities and zerospan tensile strengths, and exceptionally high handsheet densities are obtained.

Bleached slabwood: Handsheet apparent densities and refining requirements for the untreated, and xylanase and CBH treated pulps are similar one to another (9). These three bleached pulps have roughly similar fibre saturation points, relative viscosities, zerospan tensile strengths, and dried and rewetted fibre dimensions (Tables 1,2). The endoglucanase treated pulps, on the other hand, are more readily refined than the untreated

and xylanase and CBH pulps. This is particularly true for the EG-A pulp which is of low relative viscosity and wet zerospan tensile strength compared with EG-B and C (Table 1).

Unbleached and bleached thinnings: Handsheet apparent densities and refining requirements for the endoglucanase treated pulps are in agreement with their level of degradation as indicated by relative viscosity and wet zerospan tensile strength (Table 1) (1,9), [Appendix (1)]. High viscosity (and zerospan tensile index) correlates with low apparent density and high refining requirements, and low viscosity correlates with high apparent density and low refining requirements. Critical levels of viscosity and wet zerospan tensile index are very different for unbleached and bleached pulps. The EG-B treated bleached sample is the only thinnings pulp not grossly degraded by endoglucanase treatment based on handsheet apparent density values.

Handsheet tensile strength

Handsheet tensile strength by itself can be misleading since a range of kraft pulps can have similar tensile strengths but very different apparent density, light scattering coefficient, and tearing resistance (3). It is combinations of handsheet properties, rather than individual properties alone, that describe the papermaking potential of a kraft pulp. For this reason the influence of the enzyme treatments on handsheet tensile strength are evaluated with apparent density as the basis of comparison.

Unbleached slabwood: Tensile strengths for a given handsheet apparent density are marginally higher for the CBH treated than for either the untreated or xylanase treated pulps (Fig. 3). Refining requirements to reach a given tensile strength or apparent density are, however, marginally higher for the xylanase-E and CBH treated furnishes. Maximum tensile indices for a given refining input are greatest for the untreated pulp. For the xylanase treated pulps the slow development of tensile index and apparent density is explained by the loss of xylan-rich carbohydrates from fibre surfaces, decreased bonding potential and increased fibre stiffness.

The various endoglucanase treatments show decreased handsheet tensile strength and increased apparent density (Fig. 3). The magnitudes of these effects are dependent on the amount of cellulose degradation as indicated by relative
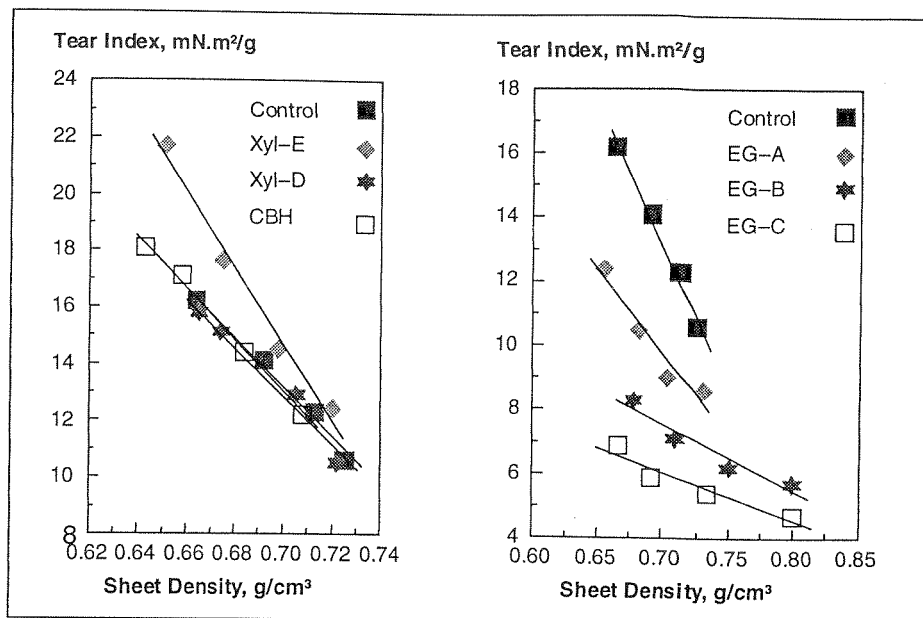
Fig. 4 Unbleached slabwood enzyme treatment, and handsheet tear index and apparent density relationships (xylanases and CBH, 400 µg/g; EG-A; B and C, 100 µg/g).
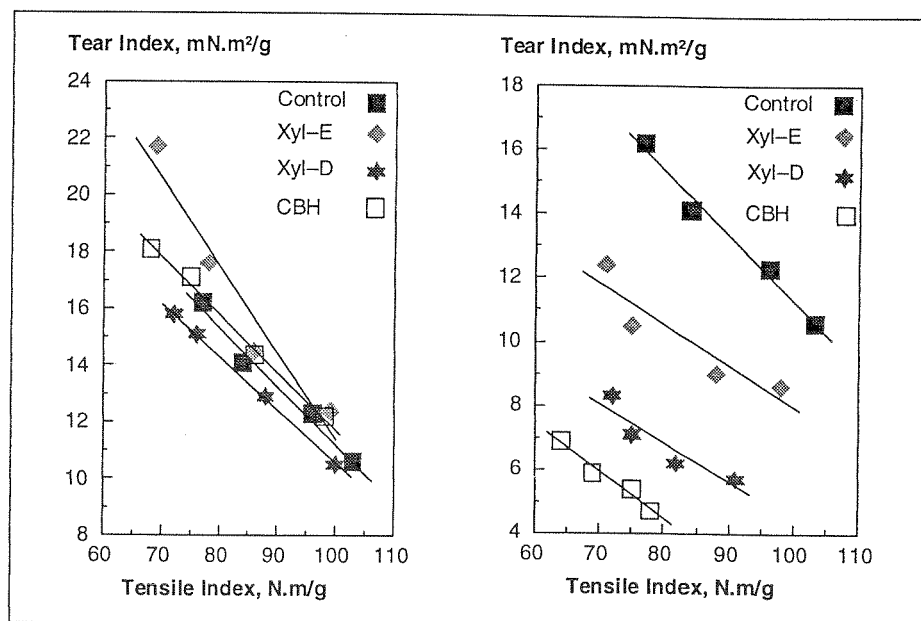
Fig. 5 Unbleached slabwood enzyme treatment, and handsheet tear index and tensile index relationships (xylanases and CBH, 400 µg/g; EG-A; B and C, 100 µg/g).

viscosity and wet zerospan tensile strength (Table 1). The EG-B 100 µg/g pulp being the least degraded shows tensile strength/apparent density relations equivalent to those of the untreated pulp. *Bleached slabwood:* Handsheet tensile strength/apparent density relationships for the untreated, and xylanase and CBH treated pulps are similar to those of corresponding unbleached pulps. The bleached pulps develop tensile strength with refining somewhat faster than the unbleached pulps.

For the endoglucanase treatments the development of tensile strength with refining is slow and out of balance with expected increases in apparent density. (9) This effect is, however, similar to that

shown for the unbleached slabwood pulps where tensile strength development decreases, and handsheet apparent density increases, in accordance with the extent of cellulose degradation brought about by endoglucanase treatment (Fig. 3). Similar effects and explanations hold for the bleached and unbleached thinnings pulps treated with endoglucanases.

Handsheet tearing resistance

Enzyme treatment effects on handsheet tearing resistance are compared with apparent density and tensile index as the bases of comparison. Out-of-plane handsheet tearing resistance is considered to be strongly influenced by the balance between fibre pull-out and fibre fracture

in the failure zone (2,13). Fibre pull-out decreases and fibre fracture increases with increasing pulp refining and fibre bonding.

Unbleached slabwood: The xylanase-E treatment is most effective in developing tearing resistance at low apparent densities (Fig. 4). This selective enhancement of tearing resistance decreases with increasing apparent density, with values similar to those of the untreated pulp being obtained at apparent densities greater than about 0.70 g/cm^3 (Fig. 4). The tear/apparent density relationship for the CBH treatment is similar to that of the untreated pulp, but able to be extended to much lower apparent density values. Tear/apparent density regressions for the untreated and xylanase-D pulps are essentially identical. Similar relationships are obtained using tensile strength as the comparison base, although the xylanase-D pulp could be marginally deficient in tearing resistance compared with the untreated pulp (Fig. 5). All the endoglucanase treated pulps are deficient in tearing resistance when compared against apparent density and tensile index (Fig. 4,5).

Interrelationships between enzyme treatments, tear index and fibre properties are explained as follows –

* The xylanase-E treatment causes a selective loss in xylan-rich carbohydrates from within fibre surfaces which lowers their affinity for water and bonding potential. Xylan is concentrated in the outermost layers of kraft fibres (12). The treated fibres also increase in stiffness as indicated by decreased fibre saturation points and decreased handsheet apparent density (Table 3, Fig. 4). Fibre strength as indicated by relative viscosity and zerospan tensile index, as well as fibre dimensions, are essentially unchanged by the xylanase treatments (Tables 1,2). Thus selected increases in tearing resistance are to be expected with the xylanase-E treatment since fibre bonding potential is decreased, fibre stiffness is increased, and fibre strengths and dimensions are unchanged (2,13). The xylanase-D treatment is non-selective in developing tearing resistance (Fig. 4), possibly because of higher wall porosity (lower stiffness) and marginally higher levels of degradation (Table 1).

* The CBH fibres retain their intrinsic strength as well as bonding potential indicated by minimal carbohydrate losses (Table 1). CBH fibres in handsheets are, however, relatively stiff and large in size since they are resistant to collapse and wall contraction during pressing and drying sheetmaking processes (Table 2) and fibre saturation points are lowered (Table 3). Furthermore, CBH treatment causes pulps to be effectively 'de-beaten' but not selectively improved since tearing resistance increases proportionately with decreasing apparent density and tensile index (Fig. 4,5). Thus enzyme treatments need to be able to modify both fibre bonding potential and stiffness to be able to selectively enhance tearing resistance for given apparent densities and tensile strengths.

* The endoglucanase treatment conditions caused gross fibre degradation as indicated by relative viscosity and zerospan tensile index (Table 1). In consequence tear index values are also low (Fig. 4,5).

Bleached slabwood: Tearing resistance for given apparent densities and tensile strengths are markedly higher for the xylanase-E than for the untreated and CBH treated pulps (9,1). The loss of xylan-rich carbohydrates from within fibre surfaces and its effects on bonding potential and fibre stiffness must again be the major reason for this selective increase in tearing resistance (Table 1). All other measured fibre and pulp properties are roughly equivalent for the xylanase-E treated and untreated pulps (Tables 1,2,3).

CONCLUSIONS

Fibre and handsheet properties can be greatly modified using selected xylanase, cellobiohydrolase and several endoglucanase enzyme treatments. Unbleached slabwood fibres of high coarseness with thick walls show the greatest response to enzyme treatment. The additional carbohydrate material and/or residual lignin present in the robust fibres of the unbleached slabwood pulp interact in some way during or after enzyme treatment. Corresponding fibre and handsheet property differences are smaller for the bleached than for the unbleached slabwood pulp, and even smaller for the thinnings pulps.

Xylanase treatments selectively dissolve xylan-rich carbohydrates and lower fibre saturation points, but retain the intrinsic fibre strengths and fibre dimensions of the untreated pulp. Thus xylanase treated fibres can be stiffer with surfaces of lower water affinity and bonding potential than untreated fibres. In consequence, handsheet tensile index and apparent density can be decreased, and tearing resistance at given apparent density and tensile strength can be selec-tively increased. Such changes in handsheet properties can be obtained without greatly increased pulp refining requirements. The xylanase-E treatment is more effective than the xylanase-D treatment in modifying handsheet and fibre properties.

Cellobiohydrolase (CBH) treated fibres retain the intrinsic strength and bonding potential of untreated fibres, but fibre saturation points are lowered. When in handsheets CBH fibres can be stiff and large in size since they are resistant to collapse and wall contraction during sheetmaking processes. In consequence, CBH treatment can cause pulps to be effectively 'de-beaten' but not selectively improved since tearing resistances increase proportionately with decreasing apparent density and tensile index. Enzyme treatments need to be able to modify both fibre bonding potential and stiffness to be able to selectively enhance tearing resistance for given apparent densities and tensile strengths.

Endoglucanase treatment at 100 μg/g can cause fibre degradation resulting in poor intrinsic fibre strengths which are reflected in handsheet properties—high apparent densities and low tensile and tearing resistance.

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Manuscript received for publication 22.7.96