REVIEW PAPER

Extraction of cellulose fibers from flax and hemp: a review

Avinash P. Manian (iD) · **Michael Cordin** (iD) · **Tung Pham** (iD)

Abstract The paper is a review on the extraction processes of cellulosic fibers from flax and hemp. The two lignocellulosic crops have a long history of use by humans for extraction of the bast fibers among other purposes. The utility of bast fibers declined over time with industrial advances and changes to the economy, but of late, with an increase of focus on environmental impact and sustainability, there is a renewed interest in these resources. The use of biomass-based resource requires an appreciation of plant anatomy and the agronomical variables in their cultivation and harvesting. This review provides an overview of these aspects as well as of the processes of retting for initial weakening of the plant structure in preparation for fiber extraction, degumming to isolate fiber bundles, and delignification.

A. P. Manian (✉) · M. Cordin · T. Pham
Research Institute of Textile Chemistry/Physics,
University of Innsbruck, 6850 Dornbirn, Austria
e-mail: avinash.manian@uibk.ac.at

M. Cordin
e-mail: michael.cordin@uibk.ac.at

T. Pham
e-mail: tung.pham@uibk.ac.at

Introduction

Flax (*Linum usitatissimum L.*) and hemp (*Cannabis sativa L.*) are among the earliest plants cultivated by humans for fibers and extracts from their seeds and flowers. The domestication of flax is believed to have occurred in the Fertile Crescent region (Fu 2011), and the earliest known evidence of flax fiber use, dating to about 30,000 years before the present time, is found in the Republic of Georgia (Kvavadze et al. 2009). Central Asia is believed to be the likely birthplace of domesticated hemp, with the earliest known evidence of hemp fiber use, dating back to about 8,000 BCE, found in Taiwan (Clarke 1999; Tourangeau 2015). Historically, both flax and hemp have been used in the manufacture of fine cloth for apparel as well as heavy-duty materials such as sailcloth, canvas cloth, sack-cloth, and cordage. Generally, it is easier to produce finer yarns from flax and therefore hemp was often preferred for technical applications (Horne 2012; Kozasowski et al. 2012; Muzyczek 2012; Sponner et al. 2005). Hemp has also been used in the manufacture of paper (Horne 2012).

The expansion in cultivation and therefore availability of cotton and jute reduced the use of flax and hemp fibers in apparel from about the middle of the eighteenth century, and the advent and increasing availability of synthetic polymers in the twentieth century reduced their use in technical applications (Horne 2012; Salmon-Minotte and Franck 2005). Flax cultivation has continued for linseed oil and other products, but from the 1930s, hemp cultivation suffered from legal restrictions in large parts of the world over fears it will encourage production of the psychoactive substance, delta-9-tetrahydrocannabinol (THC) (Bewley-Taylor et al. 2014; John 2019). However, the recent past has seen a relaxation of regulations in some regions, and even some instances of state subsidies, that has allowed for the emergence of "industrial" hemp cultivation (defined as varieties containing less than a prescribed maximum of THC, generally 0.2%–0.3% w/w) (Horne 2012; Vantreese 2002).

With increasing focus on the use of renewable biomass for the manufacture of materials, there is renewed interest in crops such as flax and hemp (Baley et al. 2020; Baley, 2019; Crini et al. 2020; Ramesh 2019; Sadrmanesh and Chen 2019; Yan et al. 2014). At present, the EU Plant Variety Database lists about 75 varieties of hemp and about 150 varieties of flax that are or may be cultivated in the region (European Commission; European Commission).

Agronomy

The different varieties of flax can be divided in two broad categories, those suited for fiber production and those for seed production. The fiber varieties are generally taller (0.8–1.5 m) than the seed varieties (0.45–0.8 m), and exhibit optimum fiber yields when cultivated in temperate climates with annual rainfall on average of 600–650 mm (Heller, 2015). The seed varieties are more resistant to hotter climates and drought conditions. In relatively cooler climates such as in Europe, fiber flax is planted in the spring (March–April) and harvested in summer (July–August) but in warmer regions, the crops are grown over the winter, with planting in late autumn and harvest in late spring (Akin 2010). Fiber varieties are planted at greater densities (2200–2800 plants per m^2) than seed varieties (1000–1200 plants per m^2), as that promotes the

growth of thinner, straighter and taller plants leading to better quality fibers (Akin 2010; Heller et al. 2015). Fertilizers (nitrogen, phosphorous, potassium, calcium and magnesium) are required to ensure good crop yields and fiber quality, and measures are needed for weed control and for protection against fungal pathogens and pests. The duration between planting and cultivation ranges between 90–180 days (Nair et al. 2014), as the growth rate and progression through developmental stages varies with both plant variety and ambient temperature—warmer temperatures promote faster development and growth (Carlson 2008). The degree of stem lignification increases with plant development and makes fiber separation from the rest of the stem (retting) progressively difficult, and therefore it is found optimal to harvest plants for fiber extraction during the flowering stage, before seed maturity is attained (Akin 2010, 2013; Meijer et al. 1995). However, that denies farmers potential supplemental income from seed production, which is a motivation for development of "dual-use" varieties yielding both fibers and seeds. An option explored is the cultivation of varieties that exhibit low degrees of lignification even at the seed maturity stage (Wrobel-Kwiatkowska et al. 2007). However, lignin acts to protect plants against pathogens and pests (Liu et al. 2018), and thus the low-lignin flax varieties may be more susceptible to infection from pathogenic fungi such as *Fusarium oxysporum* (Wrobel-Kwiatkowska et al. 2007).

Hemp plants grow to between 1–6 m in height depending on the variety and cultivation practices (Fike 2016). It grows optimally in temperate climates where the temperatures range between 15–27 °C, with annual rainfall on average between 630–750 mm (Adesina et al. 2020). However, hemp is a hardy plant and can be cultivated over a wide range of temperatures and precipitation levels (Dhondt 2020; Żuk-Gołaszewska and Gołaszewski 2020). It is also shown to be cultivable on degraded land not suitable for other crops due to problems with soil salinity and alkalinity (Zhao et al. 2021). For fiber production, the planting densities vary over a wide range (50–750 plants per m^2) depending on end use and desired fineness, whereas the densities are lower for seed production (30–75 plants per m^2) (Amaducci and Gusovius 2010; Ranalli 1999). In the open, hemp is planted in spring–summer, and the duration of plant development and

growth is strongly influenced by plant variety and environmental factors (latitude, elevation, air temperature, humidity, soil type) (Campbell et al. 2019; Ranalli 1999). It is also strongly influenced by photoperiodicity, i.e. development is accelerated by periods of short days/long nights (Amaducci et al. 2008; Fike 2016). Fertilization becomes necessary to maintain adequate levels of nitrogen especially if hemp is planted in poorly fertile soils, but as it is a fast growing plant, it can outcompete weeds and thus generally requires low levels of herbicide application (Fike 2016). However, at lower planting densities, when the canopy cover is insufficient to filter sunlight, herbicide use may be required for weed control. The harvesting of plants for fiber production (roughly 70–90 days after sowing) is preferably done at the flowering stage, as further maturation increases the proportion of "secondary" bast fibers in plants, which are shorter in length and more heavily lignified than the primary bast fibers (Amaducci and Gusovius 2010; Fike 2016). Note that secondary fibers do not occur in flax (Gorshkova, 2012). As with flax, work is underway to develop "dual-use" hemp varieties yielding maximal seed and fiber output taking into account variations in geography, climate, water availability, and agronomic practices (Baldini et al. 2020; Fike 2016; Shuvo 2020; Vandepitte et al. 2020). Another alternative is "baby" hemp cultivation, where seeds are planted at high densities in nutrient-rich soil, which encourages rapid growth of plants to acceptable heights without significant accumulation of lignin (Amaducci 2005).

Both flax and hemp may be sown as rotation crops, and it is especially recommended that flax not be grown on the same field more than once in 5–7 years to avoid propagation of fungal infestations in the soil (Heller et al. 2015; Piotrowski and Carus 2011). There is work to show that hemp may be continuously cultivated on the same field for several years without negatively impacting yield (Gorchs et al. 2017). However, planting hemp in rotation with other crops, such as cereals, offers the advantages of improving soil quality, suppressing weed, pest and pathogen infestations, and improving yields (Adesina et al. 2020; Piotrowski and Carus 2011).

Anatomy

Both flax and hemp are dicotyledonous plants, and as such, exhibit similar anatomies in their stems – from where fibers are extracted. Stem cross-sections exhibit the following entities moving outwards from the center to the periphery: pith, xylem tissue (or woody core), vascular cambium, phloem, cortical parenchyma, epidermis and cuticle (Akin 2010; Chabbert et al. 2013; Goudenhooft et al. 2019; Réquilé et al. 2018a). The "bast" fibers, which constitute the sclerenchyma tissue of the plants, develop in the phloem region as discrete bundles encircling the woody core, and grow in a direction parallel to the vertical axis of the plant. Plant growth is categorized into two types: vertical growth or lengthening of the plant, labelled "primary" growth; and lateral growth or thickening of the plant, labelled "secondary" growth. The growth occurs via division and differentiation of cells from meristematic tissue, and the procambial (or apical) meristem is responsible for primary growth whereas the cambial meristem is responsible for secondary growth.

Bundles of primary bast fibers are formed during primary growth in both flax and hemp. But only in hemp, the cambial meristem produces bundles of secondary bast fibers that begin to appear in plant sections where primary growth has ceased (Amaducci and Gusovius 2010; Chabbert et al. 2013; Mokshina et al. 2018; van Dam and Gorshkova 2003). Thus, primary fibers appear in the early stages of plant development and run through the plant length, while secondary fibers appears in the later stages and are found towards the bottom parts of stems, located in a zone between the woody core and the primary fiber bundles. Fibers also occur in the xylem tissue regions, or the woody cores (termed "shive" and "hurd" in flax and hemp respectively), but they are far shorter, more heavily lignified, and do not occur in bundles (Amaducci and Gusovius 2010; Chernova et al. 2017). The primary and secondary fibers are also termed as the "extraxylary" fibers, since both occur outside xylem tissue regions in the plant, and fibers from the woody cores are termed "xylary" fibers. (Amaducci and Gusovius 2010). Cross-sections of flax and hemp stems showing the primary and secondary fiber bundles, cambium and the xylem tissue, reproduced from literature, are shown in Fig. 1.

The primary fiber bundles, which run along almost the entire length of the plant stem, are constituted by
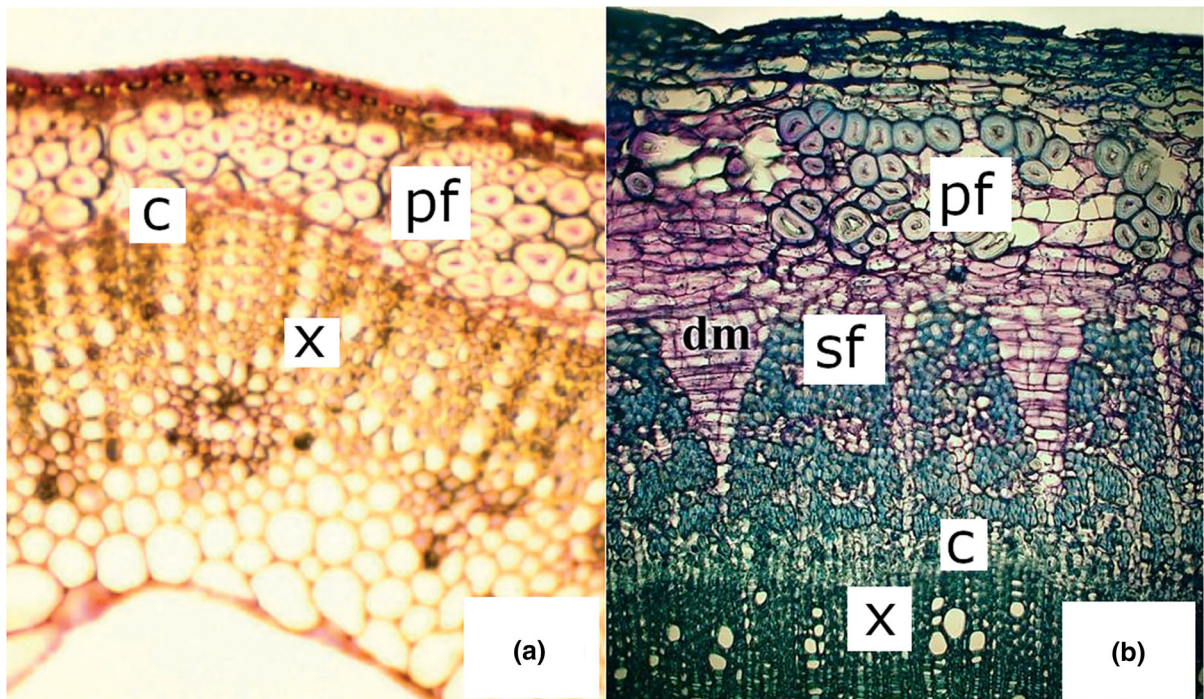
Fig. 1 Cross-sections of flax (A) and hemp (B): "pf" = primary fiber bundles, "sf" = secondary fiber bundles (only in hemp), "c" = cambium, "x" = xylem. Part (A) is reproduced with permission from D.E. Akin, "Plant cell wall aromatics: influence on degradation of biomass". *Biofuels, Bioproducts* *and Biorefining* **2008**, 2, 288–303, John Wiley and Sons. Part (B) is reproduced under Creative Commons CC BY license from A. Snegireva et al., "Intrusive growth of primary and secondary phloem fibres in hemp stem determines fibre-bundle formation and structure". *AoB PLANTS* **2015**, 7, Oxford University Press

smaller "elementary" fibers aligned together in an overlapped fashion with a slight twist, such that at any point along the bundle length, a cross-section will reveal about 10–40 elementary fibers (Gorshkova et al. 2012; van Dam and Gorshkova 2003). Primary fiber cells originate on stems at the sites of leaf traces,[1] and exhibit different stages in their growth and development (Mokshina et al. 2018). The initial growth along the stem axis (i.e. fiber elongation) occurs in tandem with other cells in the surrounding tissues, which is termed "symplastic" or "coordinated" growth (Gorshkova et al. 2018; Goudenhooft et al. 2019). That continues for some hours, until the rate of fiber elongation exceeds that of the surrounding cells, whereupon the fiber cells extend in both directions by pushing through between cells in the surrounding tissues, in what is termed "intrusive growth", which continues for several days. That results in the typical structure of fiber bundles, that of tightly packed overlapping elementary fibers. The degrees of intrusion, and thereby the thickness of fiber bundles, are sensitive to environmental stresses and thus variations

in them may be observed along the stem length depending on changing climatic conditions during plant development (Chernova et al. 2017). A schematic illustration of the intrusive growth is shown in Fig. 2.

Secondary fiber cells (which occur only in hemp) are initiated at the cambium, with the first cells appearing approximately halfway along the stem length followed by a progression in cell appearance towards the direction of the root (Chernova et al. 2017). They appear in the course of radial stem growth in sections where elongations have already ceased, and thus secondary fiber cells elongate only through intrusive growth. That, in addition to the presence of lignified mature cells in the surrounding tissues, results in secondary fiber bundles being shorter than the primary fiber bundles.

The fiber cells develop into multi-layered structures in the course of their maturation into elementary fibers. During the elongation phase, the cells consist predominantly of the Primary Cell Wall (PCW) composed of cellulose, pectins and hemicelluloses
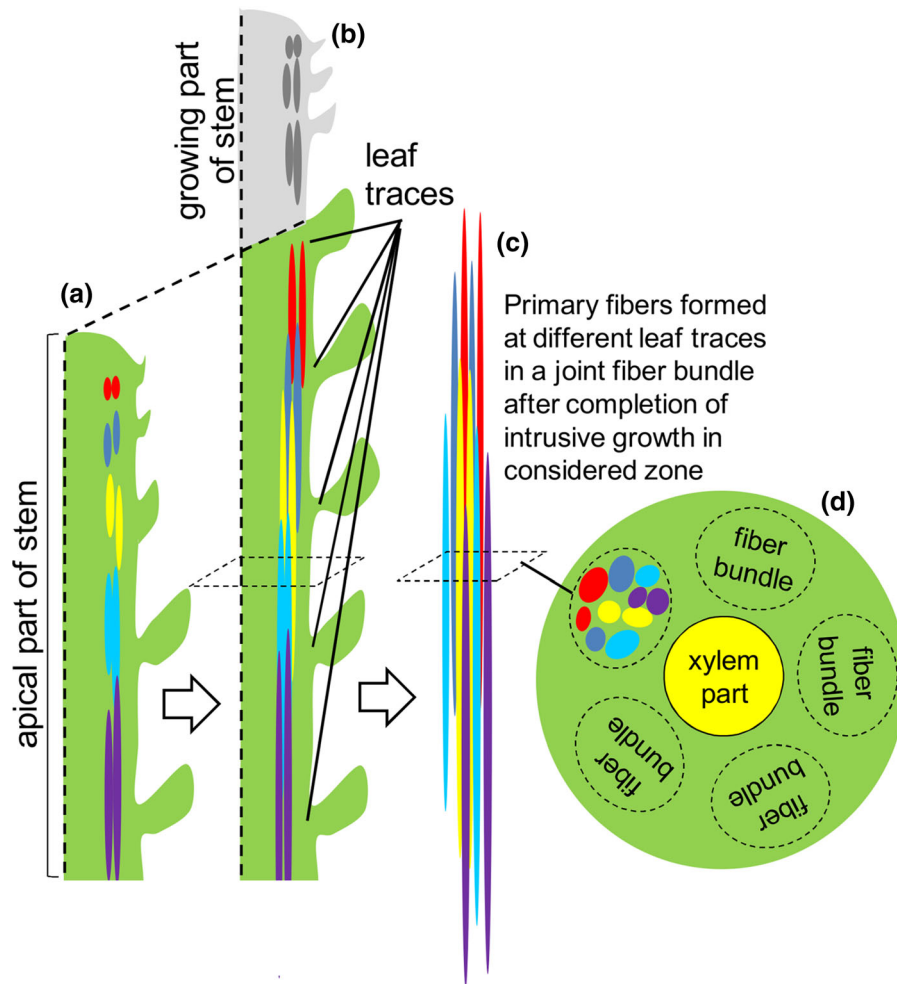
Fig. 2 Schematic illustration of the intrusive growth process. The colored spots in part (a) indicate primary fiber cells, with those initiated at different leaf traces colored differently. The spot sizes represent different stages of development, with small spots representing incipient cells and long streaks more developed cells. Part (b) illustrates the formation of a fiber bundle through elongations of individual fiber cells, and part (c) is a close-up of a fiber bundle. Part (d) is illustrative of a stem cross-section with multiple bundles encircling the woody core. Image reproduced under Creative Commons CC BY license from Mokshina, N.; Chernova, T.; Galinousky, D.; Gorshkov, O.; Gorshkova, T. Key Stages of Fiber Development as Determinants of Bast Fiber Yield and Quality. Fibers 2018, 6, 20. MDPI (Basel, Switzerland)

(Chabbert et al. 2013; Goudenhooft et al. 2019). The elongation phase is followed by cell thickening, when the Secondary Cell Wall (SCW) is deposited on the insides of the PCW over many weeks. The SCW consists of three layers, labeled S1, S2 and S3 in the direction from the PCW to the lumen at the center. The S1 is composed of cellulose, hemicelluloses, pectins and lignin, while S2 and S3 are composed of cellulose, hemicelluloses and pectins. The S2 and S3 are also referred to as the G and Gn layers, and there is an argument for the G layer to be termed the Tertiary Cell Wall (TCW) but that is under debate (Clair et al. 2018; Gorshkova et al. 2012; Goudenhooft et al. 2019; Mokshina et al. 2018). A schematic illustration of the layers in elementary fibers is shown in Fig. 3.

The cellulose is laid down in the form of microfibrils, which crisscross along the fiber length in the PCW but twist in a helical configuration around the fiber axis

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[1] Leaf traces are structures connecting the vascular systems of leaves to that of the stem, and serve in the back and forth transport of water, minerals and photosynthesis products. One leaf may be associated with several leaf traces. (Pandey 2005).
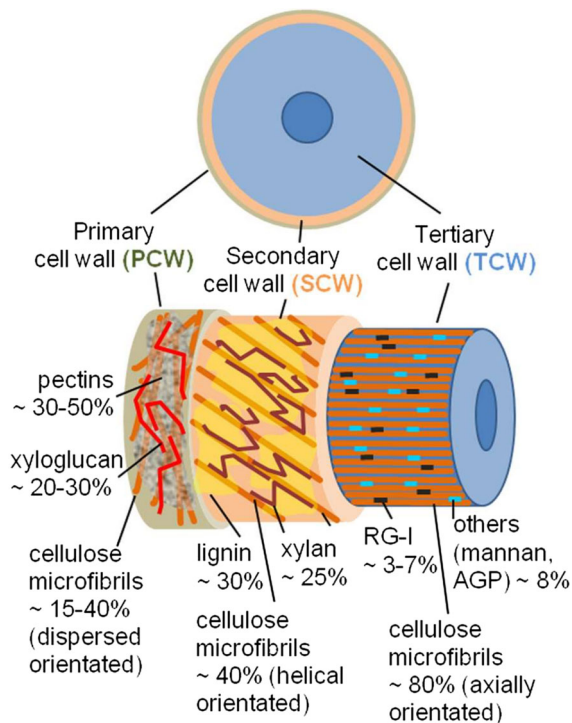
Fig. 3 Schematic illustration of the layers in elementary fibers. Image reproduced under Creative Commons CC BY license from Mokshina, N.; Chernova, T.; Galinousky, D.; Gorshkov, O.; Gorshkova, T. Key Stages of Fiber Development as Determinants of Bast Fiber Yield and Quality. Fibers 2018, 6, 20. MDPI (Basel, Switzerland)

in the SCW layers. Their angle of orientation to the fiber axis (microfibril angle, MFA) differs: in the PCW, it varies over a wide range; in the S1 layer it is in the range of 60–80° or higher; while in the S2 layer the microfibrils are nearly coaxial to the fiber with MFA of about 8–10° in flax and about 2.7° in hemp (Baley et al. 2020; Chernova et al. 2017; Goudenhooft et al. 2019). The S3 layer is essentially nascence of the S2, and often not visible in mature fibers. But when observed, it exhibits a loosely packed, heterogeneous structure. In primary fibers, the S2 layer constitutes the major proportion of the fiber bulk with the PCW and S1 layers constituting minor proportions.

The interaction forces between elementary fibers in a bundle exceed that between the bundle and its surroundings, which is attributed to (Chabbert et al. 2013; Gorshkova et al. 2012; Rognes et al. 2000):

- The shape of individual elementary fibers (narrow and elongated) promotes better interaction

between them as compared with smaller, rounded shapes of other cells in the surrounding tissue;

- Tight packing between the elementary fibers due to the intrusive mode of their growth and elongation; and,

- The middle lamella (interface between elementary fibers) contains low-methylated, highly-branched pectins allowing for strong calcium pectate gels, and deposits of phenolics including lignin.

The proportions and compositions of the main components (cellulose, pectins, hemicelluloses and lignin) vary between the different cell wall layers in elementary fibers, between primary elementary fibers from hemp and flax, and between primary and secondary elementary fibers in hemp (Chernova et al. 2018; Goudenhooft et al. 2019; Mokshina et al. 2018). A comparison of the overall features of primary fiber bundles from flax and hemp, in terms of their dimensions and chemical composition is shown in Table 1. The bundle lengths are longer in hemp compared to flax due to the differences in plant height. The lengths of elementary fibers are similar in both but their widths in hemp are larger than in flax. Hemp primary bundles contain marginally higher proportions of lignin and also contain parenchyma cells, which is not observed in flax. Further, the hemp primary bundles frequently split and merge with adjacent bundles along the stem, but that does not happen in flax where the bundles maintain unity along the entire stem length (Snegireva et al. 2015). Table 1 also shows that xylary fibers (i.e. fibers from shives and hurds) do not form bundles, are far shorter than elementary fibers in primary bundles, and are significantly more lignified.

Retting

The extraction of fibers from harvested flax and hemp stems begins with "retting" for an initial weakening of interactions between the fiber bundles and the woody core and surrounding tissue (i.e. initial loosening of the stem structure). That is followed by mechanical processes to separate the fiber bundles. The mechanical processes may be broadly classified into the following three steps (Salmon-Minotte and Franck 2005; Sponner et al. 2005).

Table 1 Range of physical dimensions and representative chemical composition of primary bast fibers and xylary fibers of flax and hemp (van Dam and Gorshkova 2003)

	Dimensions			Chemical composition[a]			
	Bundle length (cm)	Elementary fibers		Cellulose(%)	Pectins (%)	Hemicelluloses (%)	Lignin (%)
		length (mm)	width (µm)				
Flax							
Primary bundles	30–90	13–60	12–30	75	3	15	< 1
Shives	–	0.1–0.5	10–30	37	3	25	30
Hemp							
Primary bundles	100–300	5–55	16–50	70	3	15	3
Hurds	–	0.5–0.6	15–40	40	3	25	25

[a]Note that the chemical composition of fibers and bundles differ between varieties, agronomical variables, stem sections, and extraction processes – hence, the values listed here are only representative

Breaking: To break up the woody core (i.e. shives or hurds), which is achieved by crushing plant stems through pairs of fluted rolls.

Scutching: To remove the broken pieces of woody core as well as short fibers, which is achieved by passing the broken stems through pairs of rotating blades that strike and beat out the undesired material.

Hackling: To parallelize scutched fiber bundles and further remove pieces of woody core and short fibers, which is achieved by pulling the bundles through comb-like structures.

The end-product after hackling consists predominantly of individual fiber bundles, with some split into finer strands, i.e. elementary fibers. As such, this material may be used in technical applications, e.g. for cordage, composites and industrial fabrics. For construction of fine apparel, the hackled fibers generally need to be subjected to further chemical processing to separate the majority of bundles into finer, more uniform strands. Of the remaining plant mass, short fiber bundles of acceptable lengths may be used for yarn spinning, and the rest has traditionally been used as animal litter, heat insulation material, fuel, or natural fertilizer.

Retting may be achieved by traditional processes (field/dew or water), or through industrial technologies as described below.

Field (or dew) retting

Flax plants are pulled and hemp plants are cut at harvest, and laid out in oriented piles on the field (swathing or windrowing) (Desanlis et al. 2013; Horne 2012; Salmon-Minotte and Franck 2005). The pile heights and densities are maintained at levels that allow for good air circulation, and are 'turned' at regular intervals to ensure that all material is equally exposed to elements of the weather. Soil microorganisms populate the resting plants and metabolize their soft tissues, making it easier to separate the fiber bundles. With time, even the fiber bundles begin to be metabolized which reduces fiber quality, and therefore the plant mass needs to be removed from the field and processed at the optimal time. That sufficient levels of retting have been achieved is judged from changes in color of the plant material and by manually testing the ease of separating the woody core from the bark material. Machinery for mechanized harvesting and turning of flax is available commercially, but the hiatus in hemp cultivation has meant that older machinery is put back into use or machinery designed for other crops is repurposed, but manufacturers are beginning to develop new designs specifically for hemp (Desanlis et al. 2013; Gusovius et al. 2016; Salmon-Minotte and Franck 2005).

Both fungi and bacteria are active in field retting, and the degradation is reported to proceed in almost a sequential manner (Fernando et al. 2019; Liu et al. 2017). The first microorganisms to colonize the harvested mass are found to be fungal species, which are able to breach the cuticular layer with extracellular cutinases as well as by hyphal entry through damaged areas. That is followed by bacterial species that take advantage of the ingress points into the plant structure, and together with fungal species, metabolize

parenchyma cells between fiber bundles with pecti-nolytic enzymes and hemicellulases. The bacteria appear to colonize areas in close vicinity to fungal hyphal structures, utilizing them as "highways" into the plant. At later stages, there is an increase of microbial populations producing cellulolytic enzymes, which are responsible for damage to the cellulosic cell walls of bast fiber bundles observed in prolonged periods of retting. The fungal species most active in the retting process are found to be from the Ascomycota and Basidiomycota phyla, and bacterial species from the Proteobacteria, Actinobacteria and Bacteroidetes phyla, with the proportions of individual species varying with soil type and time of the year (Ribeiro, 2015). Cells with greater lignin content are less susceptible to microbial degradation, and that is one possible reason for the woody core to degrade at a much slower rate than the cortical region, but it is also thought possible that the architecture of the xylem tissue acts to limit microbial propagation through the structure (Chabbert 2020).

The advantages of field retting are that the residues may serve to enrich the soil, process costs are low, and problems of malodor are avoided (Adesina et al. 2020; Akin 2013). The disadvantages are that the fields remain unavailable for sowing of fresh crops, and it is difficult to control fiber yield and quality as the retting process is highly dependent on the weather as well as geography (Placet et al. 2017). Some alternatives that have been explored to limit influence of the weather include utilizing spring frosts to aid in the separation of fiber bundles due to formation of extracellular ice crystals in the plant (Pasila 2000), and ensiling of harvested plants to allow anaerobic retting although care is required to prevent mold growth (Gusovius et al. 2019; Martin et al. 2013).

Stand retting

Also termed as pre-harvest retting, this is a variant of field retting, where the operating principle is to spray standing crop with a herbicide—formulations of glyphosate (N-(phosphonomethyl)glycine) are com-monly employed—which then permeates through the plant via the phloem tissues (translocation) causing death and desiccation of the plants (Shekhar Sharma et al. 1989). Microbial populations are then able to colonize and degrade the plant soft tissues (similar to field retting), and aid in separation of the fiber bundles.

The advantage is that stand retting does not require swathing and turning, and the plants after retting may directly be harvested and transported for further processing. Alternatives to the spraying of herbicides include the use of open flames to terminate plant growth or allow the plants to die naturally (Assirelli et al. 2020; Ramesh 2018).

Key variables for success of this strategy is to time the spraying at the optimal growth stage of the plant, which is found to be about the mid-point of flowering (Sampaio et al. 2005). If the spraying is performed earlier, then higher proportions of immature fibers are found in the harvested plants; and if the spraying is later, then the herbicide may not be uniformly translocated and therefore the desiccation and retting levels are also non-uniform (Harwood et al. 2008; Shekhar Sharma et al. 1989). Not all varieties may yield the same response to herbicide spraying, and translocation efficacies decrease with elevation of water stress, i.e. the plants require sufficient watering for optimal results (Easson and Cooper 2002; Harvey and Crothers 1988; Harvey et al. 1985). In addition, the challenges of weather-related influence on retting progress and efficiency, as well as the field being unavailable for sowing of other crops, also apply to stand-retting.

Water retting

It describes the process where harvested plants are immersed in natural or artificial water bodies (e.g. streams or tanks respectively) to allow for microbial degradation of plant soft tissues. The retting is initiated by aerobic bacteria (from the *Bacillus* or *Paenibacillus* genus), and on exhaustion of available air, continued by anaerobic bacteria (from the *Clostridium* genus) (Di Candilo et al. 2010; Tamburini et al. 2004, 2003). The duration required for adequate retting under water (1–2 weeks) is shorter than on the field (5–6 weeks), the influence of weather and geography is minimized; and variables such as tem-perature and pH levels can be maintained at optimal levels in artificial water bodies (Magnusson and Svennerstedt 2007). Despite these advantages, and the general observation that water retting yields finer and stronger fibers, water retting has fallen from favor in Western Europe due to the costs of drying wet fiber bundles and treating the wastewater, and problems of

malodor in the retted products (Akin 2013; Jankauskiene et al. 2015; Liu et al. 2017; Morrison 2000; Morrison et al. 2000).

There are challenges with field, stand and water retting in the decision making of when to cease the process and transport the biomass to the next stage of processing. It is important to achieve control over the degree of retting, since if the plant material is retted overlong; the cellulose begins to be degraded. In field retting, attempts have been made to define "standard" days that normalize retting durations with respect to average daily temperatures and humidity (Réquilé, 2021). However, the calculation of standard days does not account for variations in precipitation levels, soil drainage and strength of solar radiation, all of which influence the rate of microbial growth and propagation, which limits the use of this approach. However, advances in microbiological and biochemical sciences, namely in metagenomics, metatransciptomics and metaproteomics, are making it possible to investigate and analyze these correlations to improve understanding and thereby achieve greater control over the retting process (Djemiel et al. 2020). Other attempts to chart the course of retting and obtain consistent results, include measurements of color change in the plant mass, measuring the emanation of volatile organic compounds and odor, analyzing changes in chemical composition, or subjecting the plant mass to standardized peeling tests (Bleuze et al. 2020; Mazian et al. 2019; Mooney et al. 2001; Réquilé et al. 2018b). Nonetheless, the practice commonly followed at present is for farmers to empirically judge the adequacy of retting by sight, touch, and smell.

Industrial processes

The challenges with field and water retting have prompted investigations on developing industrial processes to produce consistent and good quality fiber bundles. The aim of such processes is often not limited only to isolating the fiber bundles, but also to divide them into finer strands. That requires removal of the cementitious material from the middle lamella (i.e. interface between elementary fibers), which is termed "degumming". When it proceeds to the extent that individual elementary fibers may be extracted, the process is termed "cottonization"[2] (Waldron and

Harwood 2010). There are primarily three treatment modes: chemical, microbial and enzymatic.

Enzymatic treatments

The raw material for enzymatic treatment may be whole stems, bark material stripped from the woody core, or "decorticated" material obtained after a brief retting (field/water) followed by mechanical processes to separate out the woody core (De Prez et al. 2019b; Juarez et al. 2013; Liu et al. 2016). In treatments of whole stems, a prior disruption of the plant structure is found necessary to allow enzyme liquors access to the soft tissues, examples of which are hydrothermal pretreatment or mechanical crimping (Akin et al. 2004; Liu et al. 2016). Other process considerations include the ratio of enzyme liquor to plant material (kg liquor / kg plant material) as that dictates material and energy requirements and therefore process costs (Akin et al. 2000).

The natural process of retting in the field and in water occurs through microbial populations utilizing pectinases, hemicellulases and cellulases, and thus in principle, enzymatic formulations should contain the same components (Akin 2013; De et al. 2018); but in practice, it is found that pectinases are key. Polygalacturonases target the low-methylated pectins in the middle lamella region, and it appears that hydrolysis of these components is of primary importance for a satisfactory separation of fiber bundles (Akin et al. 2004; Zhang et al. 2000). The pectins are crosslinked with Ca^{2+} ions, and therefore the use of chelators during the enzymatic process or in a pre- or post-treatment step is found to significantly improve separation efficiency (Akin 2013; De et al. 2018). Ethylenediaminetetraacetic acid (EDTA) is commonly employed as chelator since the enzymes operate in a mildly acidic range (pH 4–6), and most other chelators exhibit maximum complexation ability in alkaline pH (Adamsen et al. 2002a; Akin et al. 2002, 2004; Chabbert et al. 2020; De Prez et al. 2019b). The type of polygalacturonase has an influence, as the enzyme extracted from different

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[2] Note that cottonization only denotes refining the long, coarse bast fiber bundles into dimensions (length and fineness) that allow for blending with cotton. That may be achieved only through mechanical forces, but that risks damage to the fiber and greater proportions of fiber waste.

microorganisms do not exhibit the same efficiencies (Evans et al. 2002). An alternative is the use of pectin or pectate lyases which operate optimally in the alkaline range (pH 7–11) (Bonnin et al. 2009), as that suppresses the activities of other components found in enzyme formulations such as cellobiohydrolases, and improves quality of the extracted fiber bundles (Akin et al. 2007; Alix et al. 2012; De Prez et al. 2019a).

Investigations on enzymatic retting reported in literature focus predominantly on flax, and it is found that the developed treatment protocols may not yield similar results on hemp (Akin 2013; Fischer et al. 2006). It is also the case that despite promising results and commercial-scale availability of enzymatic retting formulations, there is a lack of industrial-scale implementation of processes, attributed to a combination of factors including the process costs (Akin 2013).

Microbial treatments

These treatments involve the inoculation and incubation of microbial cultures on plant material (decorticated material, stripped bark or crushed stems) at optimal pH and temperatures for requisite durations, on plants submersed in liquid tanks or on damp plant material in sealed plastic bags. Such investigations have been performed both with bacterial cultures (e.g. *Clostridium felsineum*, *Clostridium acetobutylicum*, *Geobacillus thermoglucosidasius*) and with fungal cultures (e.g. *Schizophyllum commune*, *Rhizomucor pusillus*, *Fusarium lateritium*, *Epicoccum nigrum*) (Akin et al. 1998; Di Candilo et al. 2000; Donaghy et al. 1992; Henriksson et al. 1997; Juarez et al. 2013; Li et al. 2009; Liu et al. 2017). However, such treatments suffer from the disadvantages that the processes take long and are difficult to control, and some microbial strains may be pathogenic (Akin et al. 1998; De et al. 2018).

Chemical treatments

Chemical treatments offer the advantages of being more rapid and less expensive than enzymatic processes, but often yield more coarse fibers. A common approach is to treat flax stems with solutions of complexing agents and detergents buffered to high pH (10–11) with alkali. Examples of complexing agents include EDTA, diethylenetriaminepentaacetic acid, oxalic acid, tetrasodium pyrophosphate and sodium

tripolyphosphate; the alkalis commonly employed are NaOH, KOH or Na_2CO_3; and sodium dodecyl sulfate (SDS) is widely used as detergent (Adamsen et al. 2002b; Beltran et al. 2002; Henriksson et al. 1998; Keller et al. 2001; Rognes et al. 2000; Sharma 1988). Such formulations have also been investigated as impregnation media for steam explosion processes, where the treated stems are subject to steam under high pressure followed by a rapid decompression (Garcia-Jaldon et al. 1998; Kessler et al. 1998; Vignon et al. 1996). Treatments of harvested flax with sulfur dioxide aid in preservation of moist plant material for longer durations and increase the retting rate in subsequent enzymatic treatments, but the resulting fiber bundles are coarse and prone to contain residues of inorganic salts (Easson et al. 1998; Sharma et al. 1999). Spraying of urea and EDTA after glyphosate application increases rates in stand retting (Sharma 1986).

Physical treatments

Polar molecules interact with oscillating microwave and radio frequency radiation by undergoing rapid rotations, which generates thermal energy (termed dielectric heating) in proportion to the level of interaction. That enables a degree of selectivity as pectin is more polar than cellulose and thus heats up and is degraded at greater rates (Gregoire et al. 2019; Nair et al. 2015). Physical separations of the pectin from the cellulose are also likely. Presoaking of the harvested stalks in water is found to improve pectin removal efficiency, attributed to the plasticization of pectin (Nair et al. 2014; Ruan et al. 2020a, b). The processes are envisaged either as stand-alone treatments or as pretreatments for a subsequent enzymatic or chemical process of retting, but it is to be noted that the wastewater may present the same challenges as those from water retting (Zhao et al. 2020).

No retting

It is possible to separate the plant mass into fine strands without retting purely by mechanical forces, but that increases the propensity for inducing structural defects in fibers. They are termed "kink" bands, which are often the locus of failure under stress, and arise from disruptions to cellulose chain alignments in fiber

structures (Akin 2010, 2013; Haenninen et al. 2012). Nevertheless, it has been demonstrated that composites of equal performance may be obtained with either retted or unretted fibers (Hepworth et al. 2000; Sisti et al. 2016). However, the presence of pectic substances and hemicelluloses in unretted plant material contributes significantly to moisture absorption propensities of the biomass, which can lead to fiber swelling/deswelling induced delamination as well as increased susceptibility to fungal attack – and limits the durability of composites (Kymalainen et al. 2004; Liu et al. 2017; Pott 2002). Thus, the retting process has an impact on durability, in addition to immediate mechanical performance.

The sensitivity to moisture sorption may be reduced by chemical treatments such as acetylation, or a hydrothermal process termed the "Duralin" process, where harvested stalks are autoclaved at temperatures in excess of 160 °C, dried and subjected to dry heat at temperatures above 150 °C. (Dijon et al. 2002; Stamboulis et al. 2001). It serves to depolymerize hemicellulases and lignin, which then condense to form a waterproof resin coating on fiber bundles and improve their mechanical properties.

Delignification

The amount of lignin in fiber bundles of both flax and hemp is low (see Table 1), but due to its localization in the middle lamella, delignification treatments contribute significantly to cottonization, i.e. separating fiber bundles into their constituent elementary fibers (Akin 2013; Henglein 1969; Kiyoto et al. 2018; Rahman and Sayed-Esfahani 1979). Delignification of the woody core (i.e. shives and hurds) is also of interest to obtain fibers for the pulp and paper industry. As with retting, investigations on industrial processes of delignification have focused primarily on enzymatic, microbial and chemical modes of treatment.

Enzymatic treatments

Lignin is susceptible to the oxidative enzymes manganese peroxidases, lignin peroxidases and laccases, but laccase systems have most commonly been investigated for enzymatic delignification treatments (Fillat et al. 2010). The use of laccases alone yields limited results, as the delignification proceeds through

heterogeneous redox reactions, and the enzymes often have limited access within substrate structures. Therefore, smaller molecules are employed as mediators. The principle is that mediator molecules are oxidized by the enzyme, diffuse through the substrate structure to oxidize lignin and other aromatic structures, become reduced in that process, and thus are available to repeat the cycle (Christopher et al. 2014). Mediators also enable the oxidation of non-phenolic units of lignin, which cannot be achieved with laccases alone due to their low redox potential (0.5–0.8 V). Both synthetic molecules (e.g. 1-hydroxybenzotriazole, violuric acid) and naturally occurring compounds (e.g. syringaldehyde, acetosyringone, p-coumaric acid) have been investigated as mediators, and the most effective are found to be N-hydroxy compounds, i.e. those that contain a –N(OH)– group (Camarero, 2004; Fillat et al. 2010, 2011, 2012; Fillat and Blanca Roncero 2009; Fillat and Roncero 2009a, b). An alternative is to employ hemicellulases, as the lignin exists in complexes with xylans and mannans, and thus the destruction of these complexes releases the lignin (Cheshkova et al. 2013).

Microbial treatments

White-rot fungi, e.g. from the *Bjerkandera* and *Phanerochaete* genus, have been widely investigated for delignification treatments, as they degrade lignin at higher rates compared to other plant components (e.g. cellulose) (Dorado et al. 2001a). That selectivity for lignin degradation may be further improved by reducing nitrogen levels (or increasing the carbon-to-nitrogen ratio) in substrates, for example through pretreatment with protease enzymes (Dorado et al. 2001b; Huang et al. 2020). Other fungi investigated for delignification treatments include *Ceriporiopsis subvermispora*, which lack cellulases and thus exhibit inherent lignolytic selectivity (Akin 2008).

Chemical treatments

Many chemical treatments investigated for flax and hemp delignification derive from pulping processes and include the kraft, soda and sulfite methods (Correia et al. 2001; de Groot et al. 1995; Mustata 1994; Petrova et al. 2004). Bleaching operations common in pulping processes have also been investigated, with sodium chlorite (Pacaphol and Aht-Ong 2017), oxygen

(Danielewicz and Surma-Slusarska 2011; Kopania et al. 2012), hydrogen peroxide (Kopania et al. 2012; Pandey et al. 2019; Petrova et al. 2003), peracetic acid (Danielewicz and Surma-Slusarska 2011) and oxone (Stewart and Morrison 1996). Other investigations have looked at treatments with formic acid (de Vega and Ligero 2017), nitric acid (Shishonok and Shadrina 2006), ethanol/water mixtures (Gosselink et al. 1995), and ionic liquids (Fu et al. 2010). With a view to develop treatments that are more lignin-selective and generate lower chemical loads in the process wastewater, investigations have been performed on the use of hydrotropic reagents (Denisova et al. 2015) and with pressurized low polarity water (Kim and Mazza 2009).

Physical treatments

In investigations on air plasma treatments for delignification, a direct exposure of retted stalks to plasma discharge is found less effective as the process cannot be sufficiently controlled to prevent cellulose from being damaged. Indeed, the primary impact of direct treatments is seen to be the formation of significant cracks and cavities and a general roughening of fiber surfaces (Baley et al. 2019; Pejić et al. 2020). Indirect treatments are found more effective, where fiber bundles are immersed in mildly acidic or alkaline media and a plasma discharge is directed into the liquid close to the bundles or at air bubbled through the system (Maksimov and Nikiforov 2007; Titova et al. 2010). The reactive species responsible for delignification are identified to be ozone, hydroxyl radicals, hydrogen peroxide generated during the discharge, and the bundles are later subjected to an alkaline wash to extract the degraded lignin components. Plasma treatments in deionized water have been investigated for degumming, where a reduction in pectin and lignin contents is observed likely due to hydronium ions generated during discharge (Henniges et al. 2012; Ying et al. 2016). Direct plasma treatments of unretted stalks is found to increase the hydrophobicity of materials (Baltazar-Y-Jimenez and Bismarck 2007), which is similar to the Duralin process discussed above.

Summary and conclusion

Investigations on the extraction of fibers from flax and hemp has been regaining ground over recent years,

generally with a view to using them as fiber reinforcements in composites or in clothing (Baley et al. 2021; Manaia et al. 2019). The motivation for their use in composites is the equivalent performance but better sustainability profile compared to synthetics such as glass fibers (Bambach 2020). They also exhibit advantages over wood fibers in composites: longer fiber length (5–55 mm vs 1–5 mm); lower lignin content (about 5% w/w vs about 30% w/w); the lumen occupies a smaller area (0–5% vs 20–70%); and a lower MFA (3–10° vs 3–50°) (Madsen and Gamstedt 2013). The motivation for their use in clothing is the lower environmental impact of flax and hemp cultivation as compared to cotton (e.g. water consumption and pesticide use) (Baley et al. 2021; Möller and Popescu 2012).

Bast fiber bundles constitute only a portion of the plant dry mass (between 5–30% in hemp and 35–40% in flax) (Amaducci and Gusovius 2010; Hennink 1994; Horne 2012; Meijer 1995; Möller and Popescu 2012), and thus crops suitable only for fiber extraction may not be sufficient to sustain a viable agronomy. Hence, as mentioned above, efforts are underway to develop dual-use varieties. i.e. suitable for extraction of both seeds and for fibers.

Other investigations are directed towards developing varieties exhibiting improved tolerance to a wide range of environmental and soil factors so that cultivations may flourish in a wider set of locations thereby improving the global fiber output as well as improving local economies (Baldini et al. 2020; Goudenhooft et al. 2019; Żuk-Gołaszewska and Gołaszewski 2020). A better understanding of the molecular/genetic factors in the plant that affects fiber output and separation ease, along with improving the understanding of biotic and abiotic factors that affect microbial populations and their retting efficacies are other lines of investigation being followed (Djemiel et al. 2020; Mokshina et al. 2018; Shuvo 2020). Other efforts include investigations to establish agronomical best practices to match local soil quality and climate conditions, for example to identify optimal planting/harvesting seasons, the optimal seeding densities, and on varieties best-suited for local conditions (Baldini et al. 2020; Baley et al. 2020; Goudenhooft et al. 2019).

The global output of flax and hemp fibers as compared to other materials they can potentially

Table 2 Global output of flax and hemp fibers in 2019 (the latest year for which figures are available at the time of writing) compared to other materials for the same period

Material	Output (million metric tons)	Source
Flax fiber and tow	1.1	FAOSTAT (2021)
Hemp tow waste	0.2	FAOSTAT (2021)
Mechanical and semi-chemical wood pulp (wood fiber)[a]	35	FAOSTAT (2021)
Glass fiber	10	Thomason (2019)
Cotton	26	Opperskalski et al. (2020)
Manmade cellulosic fibers	7	Opperskalski et al. (2020)

[a] Wood fibers for composites are derived primarily from mechanical and semi-chemical pulping processes, and hence their output figures are shown (Dai and Fan 2014)

replace in composites (wood fiber, glass fiber) and clothing (cotton, manmade cellulosics) are shown in Table 2. It is possible that not all of the conventional material output is employed in composites or clothing, but even then, it is clear to see that production volumes of flax and hemp fiber are dwarfed by outputs of the other materials. There are efforts to increase the output of flax and hemp as discussed above, but it should be remembered that both flax and hemp are recommended as rotation crops, to be planted only once in every few years. It is also to be noted that in any competition for arable land between food crops vs material/energy crops, the former needs to be favored over the latter. All these factors may limit the maximal global output volumes even with the best yields. Thus, care is required when considering the end-use applications of flax and hemp fibers, with focus perhaps on niche products or those aimed at a limited geographical range, as a wholescale replacement of conventional materials with flax and hemp appears difficult.

Declarations

Conflict of interest None to declare.

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