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STRUCTURAL ASPECTS OF COTTON FIBRE MATURITY

Funded by the Cotton Research and Development Corporation under the general topic "Physical and Chemical Studies of Cotton Fibre Maturity", the project has focused on searching for links between the morphological structure of the cotton fibre and its measured maturity.

Despite a widespread, though incomplete, search of the literature; and discussions and correspondence with leading researchers here and abroad, we found no mention of a significant link. We soon learned not to be surprised, as there is much advanced work by specialists unfamiliar with related fields. For instance, an abundance of publications by botanists and plant physiologists contained no references to maturity.

We found it necessary to identify and monitor several peripheral aspects, with which we are not necessarily familiar, only acquainted. Box 1 represents these as a list, but they can be imagined as a diagram, with the nine topics surrounding and penetrating the central issue of maturity relationships. We hope that this will be helpful to other researchers having expertise in some of these fringe areas. Several other areas, notably processing, are excluded.

The Meaning of Maturity. Like other plants, cotton produces seed to provide the next generation. Approximately when the seed is "mature", it falls off the plant and may be carried by one or more natural processes. The cotton seed, exceptionally, is hairy, lending itself to transport over some distance by wind. It also follows that the fibre is more closely related to the reproductive cycle than bast fibres like jute, or leaf fibres like sisal, and may help explain why maturity is a more serious problem in cotton than in other cellulosic fibres.

In climates with windy autumn weather, this tendency to blow away must be constrained, not only with plants that resist lodging, but with bolls that only partly open, and hence must be stripper harvested. These stormproof cottons tend to be less mature, suggesting a relationship between fibre wall thickening and boll burst.

This is less relevant to Australia, where spindle harvesting predominates. That the boll burst is not simply a direct effect of fibre expansion is exploited by harvest aids based on ethephon, such as Prep. These are claimed to have no effect on micronaire, as long as the treatment guideline of four nodes between the highest cracked boll and the highest harvestable boll is followed (Rhône - Poulenc, 1992).

In the U.K. system, fibres are classified as normal, thin-walled or dead, depending on whether the mean observed wall thickness, as a fraction of the fibre diameter, is greater than 0.4, between 0.2 and 0.4 or less than 0.2 respectively. In the U.S.A., there are two categories, the criterion for wall thickness being effectively one third of total fibre width. Conversion formulas approximately link the two systems.

BOX 1: INTERFACES WITH THIS PROJECT

1. Biogenesis of cotton: plant physiology, etc. Maturation of other plants.

2. Other forms of cellulosic fibre:
   a. natural fibres, e.g., jute
   b. regenerated pure cellulose, e.g., viscose rayon
   c. forest products
   d. cultured algae, such as valonia macrophysa

3. Other fibre properties, especially those related to maturity:
   a. seed-to-lint attachment force
   b. tenacity
   c. transverse dimensions
   d. short fibre content
   e. nep formation

4. Methods of investigating structure and crystallinity:
   a. X-ray diffraction
   b. observation of fibre cross section
   c. infra-red dichroism
   d. nuclear magnetic resonance
   e. electron diffraction

5. Methods of measuring maturity:
   a. Direct microscopic examination of alkali-swollen fibres
   b. Differential dyeing
   c. Double-compression airflow
   d. Near-infra-red absorption
   e. Causticaire, i.e. micronaire after alkaline swelling
   f. Image analysis
   g. New methods under development

6. Molecular weight of cellulose:
   a. numerical
   b. weight biased
   c. estimation by fluidity

7. a. Chemical activation and reactivity of cellulose
   b. Application of (a) to dyeing and finishing

8. Genetic structural engineering of cotton fibre

9. Relationships of structural parameters and maturity with
   a. harvest aids
   b. premature senescence

To the cotton-structure scientist, maturity does not seem to have been defined. To the plant physiologist it is a measure of secondary wall growth. The outer, tubular, primary wall reaches full length about 25 days after flowering, then successive layers of cellulose are deposited inside it over about the next 40 days, the spiral direction of the chains reversing with a wavelength of about 300 μm. Depending on the interaction of the plant's heredity and the environment, especially temperature, the fibre only approaches its potential wall thickness. The maturation process ends when the plant is defoliated, either chemically or by frost. When maturation is curtailed by premature senescence (red death), limited data indicate mediocre maturity in those bolls that open.

To the fibre technologist, the mean wall thickness as a fraction of the fibre thickness is a measure of maturity. The book of Lord and Heap (1981) provides detail. In practice maturity is measured with some difficulty because a large number of fibres, many of them boderline, must be swollen in alkali, observed and classified. In cross section, a fibre has an area A - largely a function of absolute wall development - and a perimeter P - proportional to its intrinsic fineness H_s and in turn a function of variety. The relative degree of thickening, $4\pi A/P^2$, is a measure of maturity. In a circle without a hollow (lumen) this ratio is one. Empirically, the degree of thickening (θ) for a fibre having a maturity ratio (U.K.) of M=1, is 0.577; in general, $M=\theta/0.577$.

This approach is more applicable to measurement by image analysis, which is replacing labour-intensive microscopic testing, but is still slow by comparison with the other methods listed in Box 1, Section 5. We have obtained good relative, though not absolute, agreement between image analysis and known optical maturities of calibration and other cottons (Gordon, 1993).

These approaches to maturity are relevant only to the extent that they provide the mill buyer with some idea of the value and processability of a bale of cotton, other properties being equal, and a means of directing it to an appropriate end use. To the processor, maturity is that property that provides freedom from:

(a) a tendency to form neps, especially in opening and carding;
(b) excessive breakage of "dead" fibres, and a high short fibre content; and
(c) poorly dyed fibres in, e.g., denim.

The Uster manual on cotton fibre testing (Zellweger Uster, 1991) no longer has a section on maturity or fineness, but devotes seven pages to neps and their measurement. This may reflect the mix of available Uster instruments, but may also indicate future trends in fibre testing.

Processors may confuse immaturity with fineness, which can itself lead to all three problems if not carded appropriately regardless of whether the fibre is cotton, wool or manmade. The combined property is measured by airflow as "micronaire". Low-micronaire fibres, whether mature or not, are the most likely to generate neps (Griffin and Lalor, 1984). We have produced severe nepping in polyester microfibre of 90 millitex, about half the linear density of gossypium hirsutum, and a measured F.M.T. "maturity" of 1.4.

To the mill man, therefore, good maturity passes unnoticed and there is no premium for the fibre. On the other hand, immaturity is a property which:

a. Brings a discount in the cotton price.
b. Causes the card, which normally removes many of the neps that are formed in ginning and opening, to generate more neps than it takes out; this typically calls for costly resetting of clearances between carding surfaces and reduction of carding speeds.
c. Increases the rate of end-breaks, especially in spinning, again forcing reduced processing speeds

and

d. Leads to paler dye uptake and again, troublesome corrective action. These factors may combine to produce
e. White or poorly dyed neps on the surface of dyed fabrics, especially on denim and, in extreme cases,
f. "Blow-ups" in which widespread processing problems occur, with disruptions to delivery and increased production costs.

The Meaning of Molecular Weight and Degree of Polymerisation. Cotton is one of the purest forms of cellulose, which accounts for more than 90% of the dry fibre. Cellulose is the world's most widespread polymer. It has a degree of polymerisation typically exceeding 1000 in rayon and, in native cellulose, may reach 14,000 (Zahn, 1988). The "monomer" is anhydroglucopyranose, which itself has a molecular weight of 192, as $C_6 H_{12} O_6$.

This does not include a detailed discussion of the morphological structure because

1. this has been amply described elsewhere, with new information continually added to the literature;

2. little of the technology applied to textiles of cotton and other fibres derives from direct application of the abundant fundamental research on fibre structure;

3. we are concerned here with degree of polymerisation, its measurement and its possible links to the development of maturity.

This is expressed, inter alia, as:

1. number average DP, as obtained by, e.g., osmotic measurements
2. weight average DP, as obtained by, e.g., light scattering
3. Viscosity average DP, approximately a weight average DP, a further explanation of which is available (Krässig, 1985). Because of the available technology for viscometry of cellulosic fibres, this was the experimental method chosen for the present project.

As the fibre post anthesis begins with glucose and becomes a high-molecular-weight polymer as the fibre wall thickens, we might postulate a quantitative relationship between maturity and molecular weight, the latter expressed as viscosity in solution.

Peter and Bowes (1988) observed, without supplying evidence, that as the fibre matures, the cellulose content increases, while the other proportions, of pectin, protein, ash and organic acids, decline.

The two stages, of initial fibre lengthening and the thickening of the secondary wall, are not as separate or sequential as some textbooks would indicate. Evidence for their overlap is the reduction in fibre length when Australian cottons are produced under dryland conditions.

The Meaning of Crystallinity. The level of crystallinity attained in salts, some minerals and even semi-conductors is not approached in natural fibres. Man-made fibres, too, are imperfect crystals, with measured tenacities several times lower than those of the constituent chemical bonds. Such notions as the fringed-fibril theory have been applied to explain the merging of crystalline into amorphous parts.

Most contemporary results on crystalline structure of cellulose are consistent with the classical Meyer-Misch (1937) model based on x-ray diffraction. Dobb, Fernando and Sikorski (1979) confirmed this model using electron diffraction and concluded that "all the cotton types examined possessed an identical crystalline structure". They continued the investigation by examining particles left after hydrolysis in sulphuric acid, showing them all to have a width of 8 nm and normally distributed lengths averaging 75 nm.

The flexibility demanded of textile fibres - as distinct from many fibres used in fibre-reinforced composites - would be impossible in pure crystals. The amorphous parts also contribute extensibility and are richer in accessible sites for the rapid uptake of moisture and dyes.

In research stimulated by the problem of immaturity in marginal high-plains growing areas, Haigler (1992) examined the effects of coolness, especially at night, on cotton growth and maturity. Each variety was found to have a potential maturity end-point, which could not be altered by enhancing the growing medium, but which may be reached only in suitable growing conditions. The onset of secondary wall synthesis could be delayed by up to six days in vitro, and approximately so in the field. Cool nights are associated not only with immaturity but with rings in the secondary wall. So far, no means has been found of isolating these rings for structural testing.

The same team, Roberts *et al.* (1992), showed that as the minimum temperature in the cycle was increased from 15 to 28°C, respiration (using carbon 14) became normal immediately but the rate of cellulose synthesis and fibre elongation did not recover for several hours. Their research did not include tests of maturity.

Much of our understanding of the development of this crystallinity is due to the work of Deborah Delmer (1977). Discussions at Melbourne University (Bacic, 1992) centred on the role of (1→3)-β-D-glucan (callose) as an intermediate in cellulose production, as well as a product of abscission and defoliation, but led to no links to fibre maturation. The paper of Meier et al. (1981) is recommended for further reading.

Orientation and Crystallinity. In common with other fibres, cotton has both crystalline and amorphous parts. Most observers now consider that the same molecule can pass through both crystalline and amorphous regions. These properties are readily controlled in man-made fibre production, chiefly by drawing, which has the effect of orienting them longitudinally until they are sufficiently parallel to form crystallites.

Access of water and water-borne molecules to chemical bonds in cellulose is faster in amorphous than in crystalline parts. Further, reactivities of the hydroxyl groups in cellulose depend on position, the group in the six position being most likely to bond or undergo reactions such as acetylation (Fig. 1).

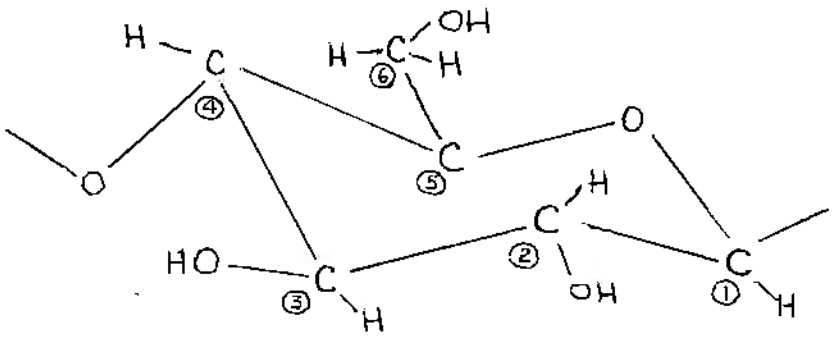

Figure 1. The Unit of Cellulose
Showing the Reactive Hydroxyl Groups.

Much of the work on the relationship between these two properties has been in the general area of chemical treatments, notably mercerisation, which causes a transition from Cellulose I to Cellulose II. Gruber (1979) described the changes in the unit cell of the crystal lattice, notably a distortion of the opposite angles from 83° to 63°. Other workers have postulated Cellulose III and IV, but the existence of these forms is not unanimously accepted, and their importance is in regenerated cellulose (rayon), not natural fibres.

Hindeleh (1980) was among the first to detect differences between the crystallinities of cotton varieties. This was followed by the work of Iyer et al. (1981) which examined 30 varieties varying in crystallinity from 74 to 87%. They also tested length, fineness and maturity, and correlated several pairs of properties. The link between crystallinity and maturity was apparently incidental to their study, for they did not include the outcome. Using their data on 29 cottons, we calculated the correlation coefficient at 0.71, with a 95% confidence interval of 0.49 to 0.86. That is, about 0.71^2 or half the variation in maturity is explained by crystallinity. This result proved to be exceptional.

Correspondence with Josef Perel (1992) yielded several opinions based on his considerable experience with x-ray diffraction in cellulose. He found some correlation between maturity and crystallinity within a variety but insisted that "any attempt to correlate crystallinity with maturity between different varieties is doomed to failure". Earlier work was presented at a Bremen Conference (Perel, 1986). He also pointed out that molecular chain length develops in the first month after anthesis, well before crystallites have reached their final form and maturation proceeds.

Experimental. We chose eight USDA calibration cottons, omitting the asiatic F cotton and the "I" sample, which is similar to "A". This modest number was due to the very high testing costs, but the values of calibration cottons are that their maturities are known with some precision, and that any other scientists can repeat our experiments and check our results.

1. Fluidity, the inverse of viscosity, is defined here as the rate at which a fluid passes through a standard orifice under its own weight. The test, developed by the Shirley Institute initially for rayon, provides a measure of the mean molecular weight.

   The untreated samples were each weighed and separately stirred with cuprammonium hydroxide solution having an excess of ammonia. This solution was poured into Shirley viscometer tubes for high molecular weight fibre and the time to reach the lower graduation was recorded. This was then converted to standard units of fluidity.

2. **Crystallinity.** Reliable measurement was beyond our expertise and apparatus, so we contacted Dr. Veronica James of the School of Physics at the University of New South Wales. The contract was taken up by Emeritus Professor Jack F. McConnell for a reasonable fee of $2000. This required some improvising to exploit their existing X-ray diffraction apparatus. Not fully satisfied with the first set of results, McConnell ran a second set. We presented both sets of results equally, thus having 16 readings and a means of comparing within-variety and between -variety variance.

   By way of completeness, and as a guide to other researchers in this field, extracts from McConnell's correspondence, in chronological order, follow:

"In the first place, my original plan to expose them at the "Photon Factory" in Japan was soon discarded because the low-angle facility that we use there cannot "see" anything much less than 15 Angstroms. I had no idea that the cotton unit cell is only a few Angstroms. We have been looking at spacings up to 3,000A. Next, I had to find a suitable X-ray diffractometer, which turned out to be computer-controlled. This is great for routine work, but a bit of a pain in the neck for an unusual job. Then I had to devise a technique which ensured that the results were not affected by the total mass of sample irradiated, since it was practically impossible to ensure this, and to ascertain whether the results were the same for different orientations of the fibres with respect to the plane of the incident and scattered X-ray beam. Having satisfied myself that I had all of these under control, I then devised a specimen holder for the specimens, and gave it to the workshop to make. Merely obtaining a trace by continuous scan gave me approximate values, but I soon realised that I needed a large number of counts to get any reliable results. I feel reasonably happy with the results, but some samples were a little short of volume, particularly specimen G, the immature one, and I had to count for a very long period to be sure of its accuracy. On an absolute scale, I would not like to claim any more than about 2% accuracy, but I think that the relative values are much more accurate. If you need any better absolute accuracy, I think that I could devise a suitable technique, but it would require extensive testing. Ideally, the specimen should be compact over a rectangular area, with a plane upper surface. Using a thin layer of tape on the top would ensure this, but this would require extensive testing to ensure that the tape did not disturb the measurements.

"I have struck a spot of trouble with the diffractometer. I have experimented quite a bit with different arrangements of the samples, and feel sure of being able to get consistent and reliable results. However I noticed that the total number of counts recorded for, say, a 5-minute exposure were considerably different at different times. Suspecting that my sample preparation might have been at fault, I carried out repeats using identical samples, and found the same differences occurring. Then I noticed that these changes in total counts occurred at intervals of a few hours, I suspected that this effect was due to changes in the X-ray generator output. Enquires of Prof. Ward, who is in charge of the Laboratory, confirmed my suspicions. It appears that the output remains constant for quite long periods, and then changes suddenly. Unless such change occurs during an actual measurement of the two required values (002 peak and amorphous) for any one specimen, no serious error in result should occur, although lower total counts would lead to higher statistical errors. Repeat measurements have satisfied me that no serious errors have occurred, but I decided to repeat a number of measurements to make certain.

"I have therefore repeated all of my measurements, in some cases many times, and now feel confident that these figures are as reliable as I can obtain without chopping the cotton into such small lengths as to enable them to be packed down to a level surface as is done with powder samples. There is, in my opinion, a fundamental weakness in the definition of the Crystallinity Index in so far as measuring it is concerned. By its very definition, the figures obtained are heavily dependent upon the measured value of the scattering from the amorphous portion of the structure, but this is by far the more difficult of the two measurements, since there is no simple way of knowing at what angle it should be measured. One paper suggests 18 degrees in 2-theta, but another says "about 19 degrees", and the figures at these two angles are sufficiently different as to change a calculated index from, say, 82% to 80%. In these last measurements, I have used 18 degrees, the value obtained for the amorphous contribution by those who progressively bashed the staples into a powder. I suspect that some people take the minimum, which is easier, but is closer to 19 degrees. It is hard to feel satisfied with any particular strategy, since around 18 degrees one is getting the effects of the "tails" of the 002 and 101 reflections on either side, and their extent depends on many factors, not the least of which is the diffractometer geometry. For the above reasons, I would not like to guarantee the absolute values given, but their relative values should be reliable"

3. Maturity. We measured micronaire, fineness and maturity of the eight cottons using the Shirley double-compression Mark 1 FMT tester. Results were calculated from Shirley formulas and expressed as means of four samples. A value of 1.0 represents full maturity; the possible range of (U.K.) maturity ratio is 0.2 to 1.2.

Results.

The information is presented graphically in Figs. 2-4, showing the extent of correlations. Fig. 2 illustrates a weak relationship between fluidity and maturity. The low coefficient, $+.14 \pm .66$ reverses to -.64, with a confidence interval -0.92 to +.11, when the most immature cotton (G) is omitted.

These are not significantly different from the results of Iyer et al, or of Ghosh.

Figure 3 describes the change of crystallinity with maturity. Both readings of crystallinity were included. The point marked "2" represents two coinciding readings. The low correlation is expressed as $+0.13 \pm 0.48$. If the two readings on the immature, and arguably non-commercial, cotton are omitted, the correlation is -0.40 ± 0.45.

Figure 4 relates the two dependent variables with each other, showing no significant relationship.

Conclusions

The project has not found congruence in the mechanisms governing

a. the development of maturity in cotton fibres
b. the degree of crystallinity in cotton
c. molecular weight, to the extent that fluidity measures it.

Our concurrent work on fungal attack ("cavitoma") on cotton has likewise indicated some paradoxes: the literature shows that affected cotton has lower measured fluidity and higher measured maturity. Clearly, the mechanisms of fungal attack also are not fully understood.

We cannot opine that either of these two measures can be used as an alternative to maturity, despite uncertainty about both the measurement and definition of maturity. Nevertheless the project has been useful in refining techniques, enlarging our competence, developing a world-wide network of contacts and forging links with related areas.

As for the future, our empirical approach may have reached its limits, and we are proposing no further activity on our part. This pilot experiment has achieved nine of the ten objectives listed in the application. If this topic is to be investigated further, it will probably need to pass to botanists who are thoroughly familiar with the phytogenesis of cellulose, the modern Australian cotton industry and the commercial significance of maturity.

REFERENCES

Bacic, A., 1992. *Personal Communications.*

Delmer, D.P., 1977. *The Biosynthesis of Cellulose and Other Plant Cell Wall Polysaccarides.* In F.A. Loewus (Ed.): *Recent Advances in Phytochemistry,* Vol. 11, p.45-48.

Dodd, M.G., L.D. Fernando and J. Sikorski,1979. *The Molecular Architecture of Cotton Cellulose.* J. Text. Inst., 68, 479-486.

Ghosh, S., 1989 or later. *Determining Cotton Fibre Maturity Using a Near Infra-Red Spectroscopic Method.* No reference; paper available on request.

Gordon, S.G., 1993. Ph.D. Thesis, La Trobe University, in preparation.

Griffin, A.C. and W.F. Lalor, 1984. *The Effect of Fibre Bundle Strength on Short Fibre Content and Nepping Potential,* 4 pp. International Cotton Conference, Bremen.

Gruber, E., 1979. *Microgel Particles in Solutions of Cellulose and Cellulose Derivatives.* Cellulose Chem. Technol., 13, 259.

Haigler, C.H., 1992. *Personal communication.*

Hindeleh, A.M. 1980. *Crystallinity, Crystallite Size and Physical Properties of Native Egyptian Cotton.* Tex. Res. J., 50, 667-674.

Iyer, P.B., K.R.K. Iyer and N.B. Patil, 1981. *Crystallinity of Native Cotton - Does it Influence Other Fiber Properties?* Tex. Res. J., 51, 679-681.

Johnson, N.A.G., 1991. *Cavitoma in Australian Cotton,*4 pp. - unpublished, copy available on request.

Krässig, H., 1985. *Cellulose.* In Ullman's Encyclopedia of Industrial Chemistry. Vol. A5, p. 377.

Lord, E. and S.A. Heap, 1981 revised. *The Origin and Assessment of Cotton Fibre Maturity.* International Institute for Cotton, 27 pp.

Meier, H., L. Buchs, A.J. Buchala and T. Homewood, 1981. *(1→3)-β-D-Glucan (callose) is a probable intermediate in biosynthesis of cellulose of cotton fibres.* Nature, 289-824.

Meyer, K.H. and L. Misch, 1937. Helv. Chim. Acta, 20, 232.

Perel, J.P., 1992 July 9. *Personal Communication.*

Perel, J.P., 1986. *Measurement of Crystallite Size in Cotton by x-ray Method; Relation to Maturity*. International Cotton Conference, Bremen, 4 pp.

Peter, U. and O. Bowes, 1988. *Quality Variations in Cotton and their Effects on Bleaching, Dyeing and Finishing*. International Cotton Conference, Bremen.

Rhône-Poulenc, 1992. *"Prep 720 has no effect on Micronair"*. Advances in Cotton, Edition 3, March, p.1.

Roberts, E.M., N.R. Rao, J.Y. Huang, N.L. Trolinder and C.H. Haigler, 1992. *Effects of Cycling Temperatures on Fiber Metabolism in Cultured Cotton Ovules*. Plant Physiol., 100, 979-986.

Segal, L., J.J. Creely, A.E. Martin and C.M. Conrad, 1959. *An Empirical Method for Estimating the Degree of Crystallinity of Native Cellulose using the X-ray Diffractometer*. Tex. Res. J., 29, 786-794 (1959).

Zahn, H., 1988. *Latest Findings on the Microstructure of Cotton*. Proc. International Cotton Conference, Bremen, 9 pp.

Zellweger Uster, 1991. *Measurement of the Quality Characteristics of Cotton Fibres*. Uster, 31pp.

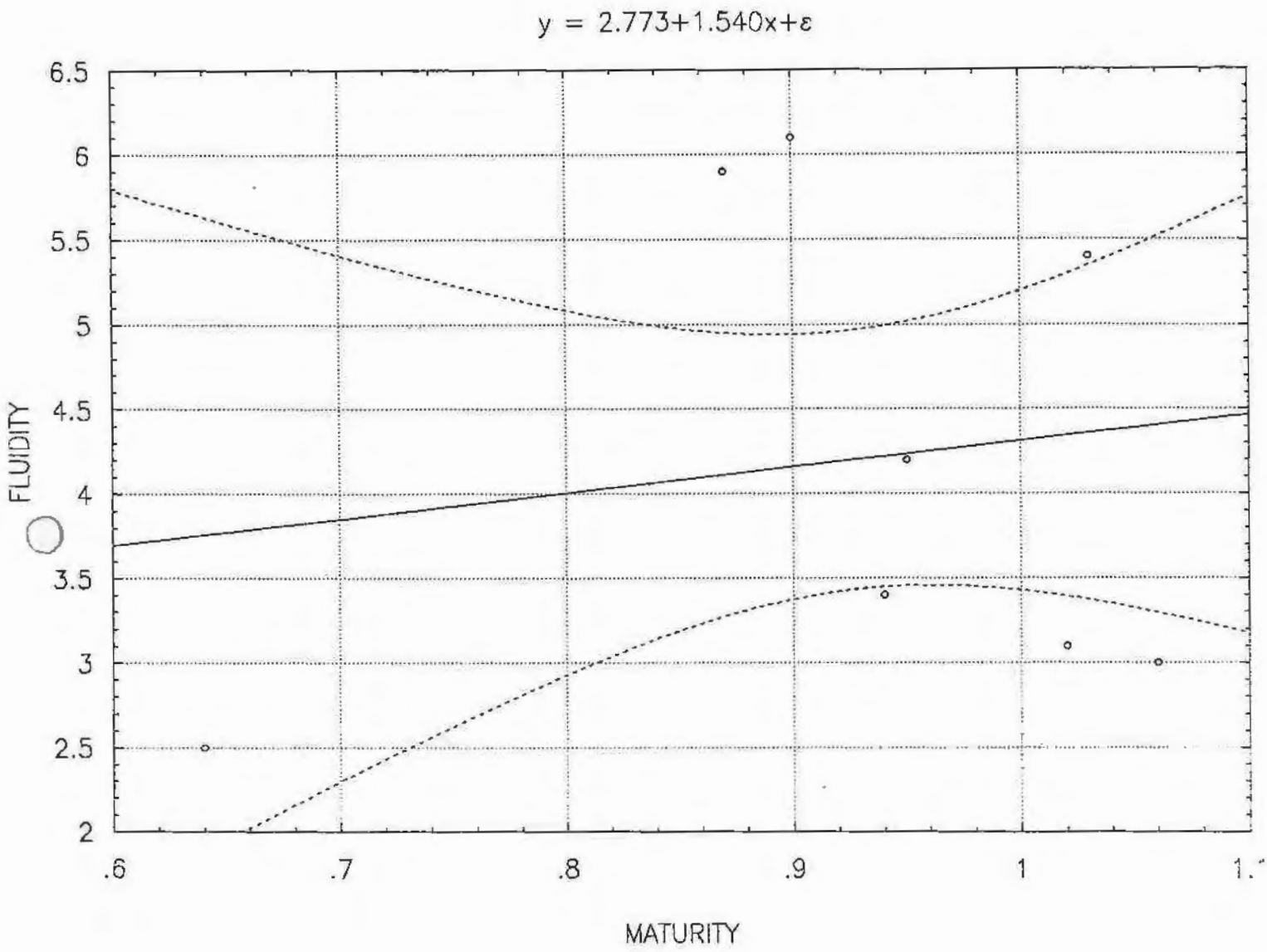

FIGURE 2. Fluidity as a Function of Fibre Maturity

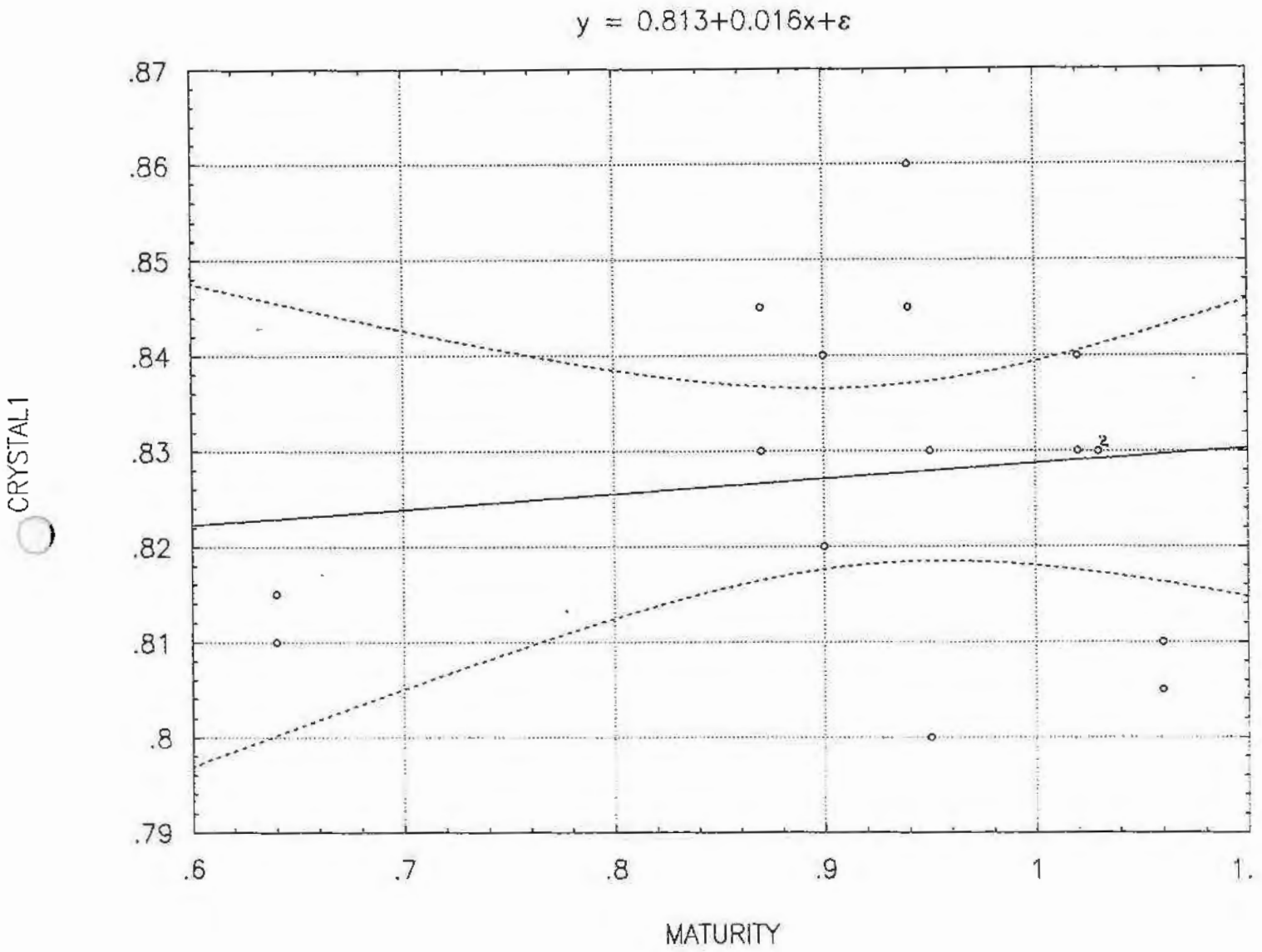

FIGURE 3. Crystallinity as a Function of Fibre Maturity

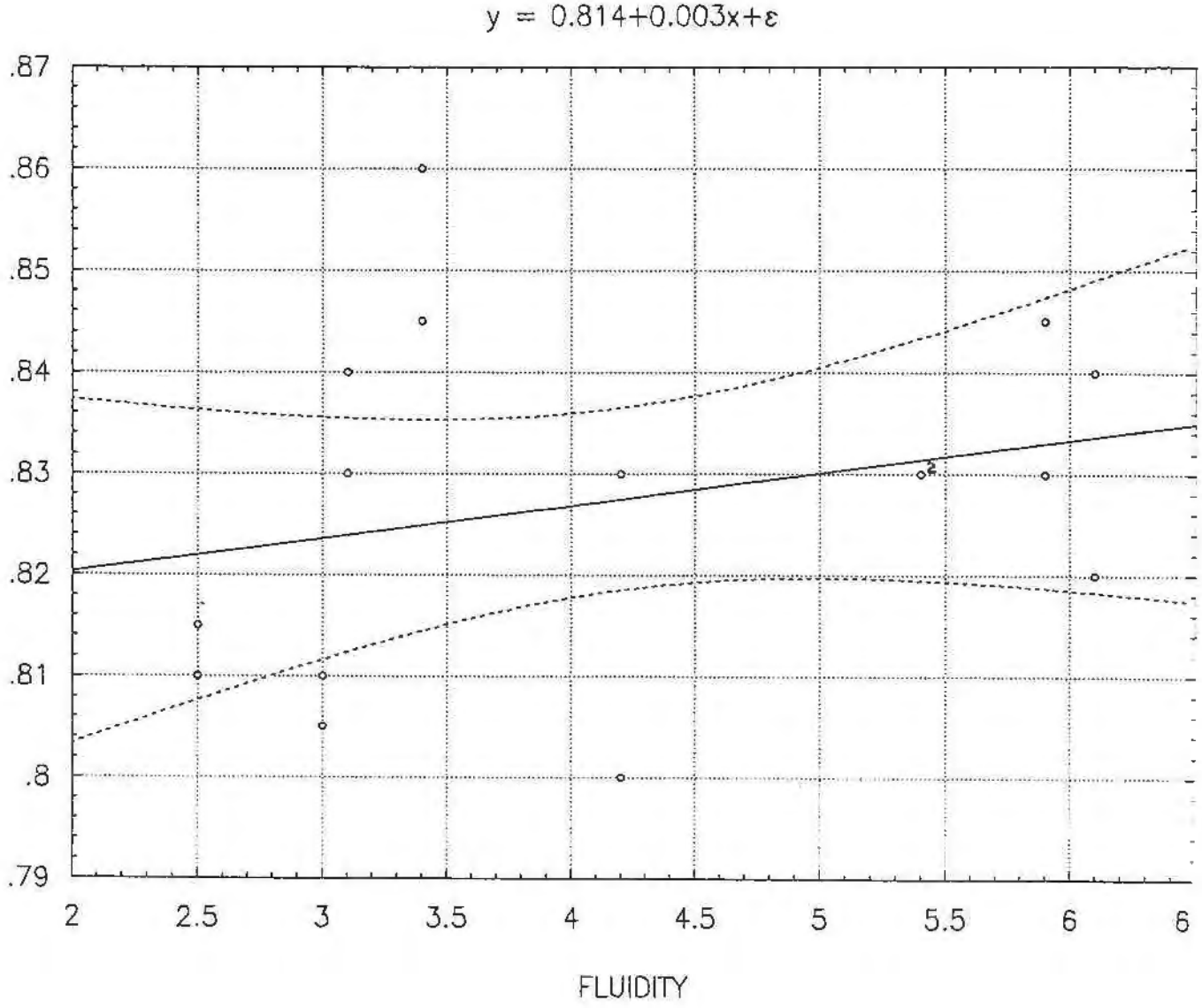

FIGURE 4. Crystallinity as a Function of Fluidity