# Possibilities of near-infrared spectroscopy for nondestructive determination of some technological properties of *Bombyx mori L.* cocoons

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## Introduction

Raw silk quality is known to be largely dependent on the cocoon properties. Some of the cocoons' parameters are important for the breeding, while some are important for reeling cocoons. Cocoons must be classified before the unravelling process. Determinations of cocoons' technological parameters are destructive and time-consuming. Fast and reliable methods for *in-vivo* analyses of cocoons are highly desirable.

Near infrared (NIR) spectroscopy has been examined as reliable method for biological material investigation. The advantages of this method include speed and simultaneous non-destructive measurement of a number of constituents. The short-wave near infrared region from 700 nm to 1100 nm is suitable for on-line measurements with fibre-optic probes. Several authors have reported applications of NIR spectroscopy for differentiation of male, female and dead silkworms while in the cocoon, and for determination of degumming loss during degumming process and sericin content in solution.[1,2] FTIR, NIR and Raman spectroscopy have been shown to be a rapid and effective ways to investigate quantitatively, as well as qualitatively, the conformation of *Bombyx mori* silk protein.[3-6]

The objective of this study is to investigate the possibilities for non-destructive determination of some technological properties of cocoons by NIR spectroscopy.

## Materials and methods

A total of 196 univoltine F1 hybrid cocoons - population of interbreeding 1013 × 1014 were used in the study. Rearing of the silkworm (*Bombyx mori* L.) was conducted during May-June 2008 in the experimental training base of the Faculty of Agriculture, Trakia University, Stara Zagora by standard rearing technique. Single cocoon reeling was carried out. Several technological parameters—filament weight, cocoon shell weight, reelability, Shell ratio and Silk ratio were determined.

NIR measurement was performed with the FQA-NIR Gun (Shizuoka Shibuya Seiki, Hamamatsu, Japan) in the wavelength region 588–1092 nm using the interactance mode. A commercial program Pirouette 2.0 (Infometrics, Inc., Woodinville, WA, USA) was used for performing of spectral data processing. PLS regression was used for quantitative analysis. Only smoothing was used for spectral data pre-treatment. The coefficient of multiple regression ($R$), the standard error of cross validation ($SECV$) and standard error of calibration ($SEC$) were used to evaluate the calibration equations.

## Results and discussion

Average second derivative spectra of tested cocoons are shown in Figure 1.

Observed absorption bands of cocoons were connected mainly with C-H 5th and 4th overtone vibrations in the region from 600 nm to 800 nm and with N-H second overtone vibrations from aromatic amines in the region from 1010 nm to 1060 nm.

Range, mean, standard deviation and NIR calibration statistics for determination of tested parameters are shown in Table 1.

The results showed a high relationship between spectra of cocoons in the short-wave NIR region and their technological parameters. Multiple correlation coefficients of calibration equations for determination of cocoon shell weight, reelability, shell ratio and silk ratio tested cocoons

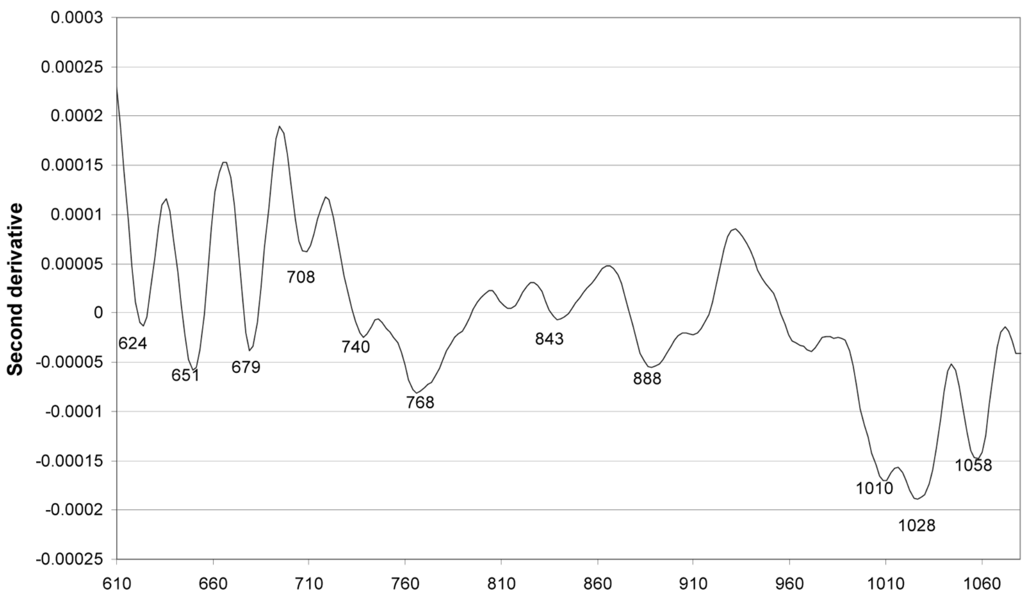


**Figure 1.** Average second derivative spectra of cocoons.

**Table 1.** Range, mean, standard deviation (*SD*) and NIR calibration statistics for determination of tested parameters.

| Parameter | Min. | Max. | Average | *SD* | NIR calibration statistics | | |
|---|---|---|---|---|---|---|---|
| | | | | | *SECV* | *SEC* | *R* |
| Filament weight, mg | 127 | 371 | 244.94 | 35.46 | 21.81 | 18.32 | 0.82 |
| Cocoon shell weight, mg | 176 | 412 | 276.03 | 37.34 | 21.20 | 12.00 | 0.93 |
| Reelability, % | 52.9 | 94.9 | 88.75 | 5.11 | 2.75 | 1.81 | 0.94 |
| Shell ratio, % | 36.9 | 63.0 | 47.24 | 4.35 | 2.05 | 1.41 | 0.93 |
| Silk ratio, % | 25.9 | 54.9 | 41.89 | 4.19 | 1.94 | 1.43 | 0.91 |

parameters were bigger than 0.91 and ratio of *SD/SEC* were bigger than 2.8, respectively. Figures 2 and 3 graphically illustrate the relationships between actual and NIR predicted values of cocoon shell weight and silk ratio, respectively.

The most important spectral information for filament weight determination in the calibration equation obtained were found to be at 645 nm, 699 nm, 710 nm, 781 nm, 880 nm, 1012 nm and 1060 nm, and for cocoon shell weight at 621 nm, 645 nm, 667 nm, 710 nm, 781 nm, 880 nm, 1017 nm, 1037 nm and 1060 nm, respectively. Therefore determination of filament weight and cocoon shell weight was based on similar spectral information. Different spectral information was

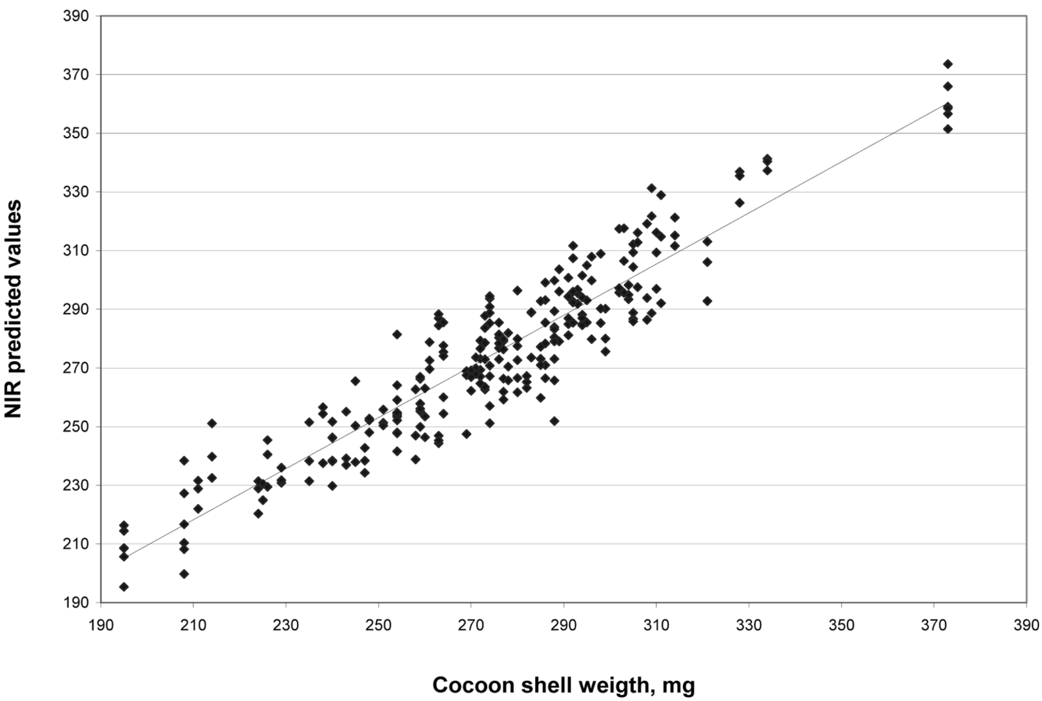


**Figure 2.** Relationship between actual and NIRS predicted values of cocoon shell weight.

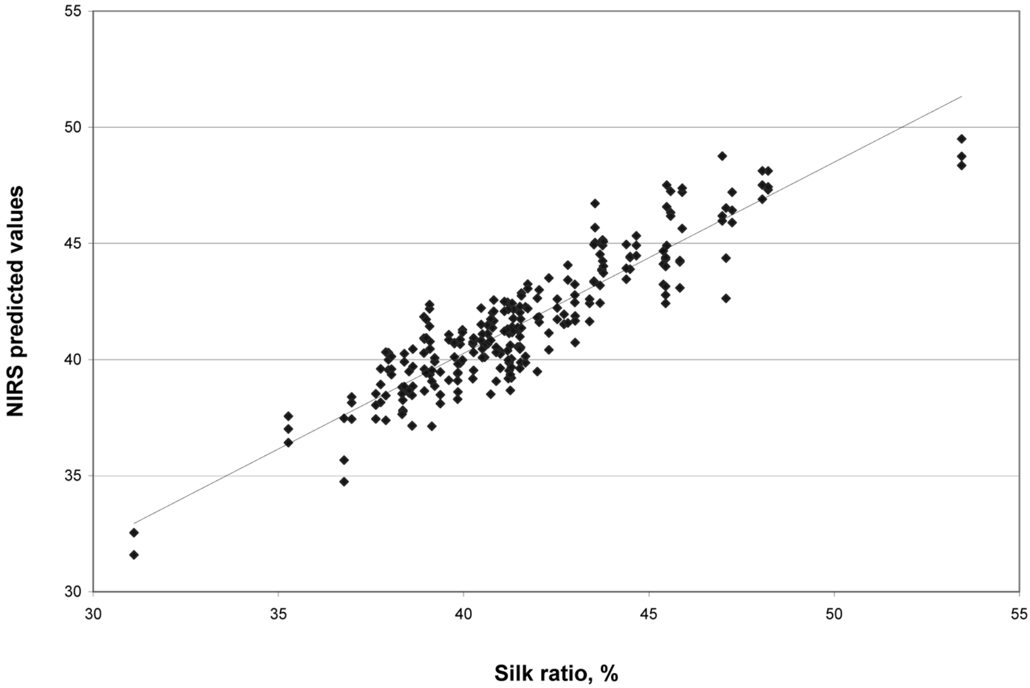


**Figure 3.** Relationship between actual and NIRS predicted values of of silk ratio.

used in equations for determination of other tested parameters. The most important wavelengths in a equation for reelability percent determination were 657 nm, 728 nm, 840 nm, 943 nm, 1017 nm, 1036 nm and 1064 nm, for shell ratio 641 nm, 683 nm, 776 nm, 882 nm, 950 nm, 1012 nm, 1028 nm and 1058 nm, and for silk ratio 645 nm, 688 nm, 780 nm, 876 nm, 1010 nm, 1056 nm, respectively.

## Conclusion

The results indicated that NIR spectroscopy showed good accuracy of determination of tested technological parameters of silk (*Bombyx mori* L.) cocoons and had the capability for fast and non-destructive quality evaluation of *in-vivo* cocoons.

## References

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