

## Prediction of Glucose Concentration in a Glucose-Lactose Mixture Based on the Reflective Optical Power at Dual Probe Wavelengths

Song Gao, Wenjing Yue, and Sang-Shin Lee\*

*Department of Electronic Engineering, Kwangwoon University,  
20 Kwangwoon-ro, Nowon-gu, Seoul 01897, South Korea*

(Received September 17, 2015 : revised November 11, 2015 : accepted December 8, 2015)

An enzyme-free optical method is proposed for estimating high concentrations of glucose in a glucose-lactose mixture, based on a predictive equation that takes advantage of the reflective optical power observed at two discrete wavelengths. Compared to the conventional absorption spectroscopy method based on Beer's Law, which is mainly valid for concentrations below hundreds of mg/dL, the proposed scheme, which relies on reflection signals, can be applied to measure higher glucose concentrations, of even several g/dL in a glucose-lactose mixture. Two probe wavelengths of 1160 and 1300 nm were selected to provide a linear relationship between the reflective power and pure glucose/lactose concentration, where the relevant linear coefficients were derived to complete the predictive equation. Glucose concentrations from 2 to 7 g/dL in a glucose-lactose mixture were efficiently estimated, using the established predictive equation based on monitored reflective powers. The standard error of prediction was 1.17 g/dL.

**Keywords :** High glucose concentration, Correlation coefficient, Glucose-lactose mixture, Reflective optical power

**OCIS codes :** (280.4788) Optical sensing and sensors; (120.5700) Reflection; (330.1880) Detection; (350.4800) Optical standards and testing

### I. INTRODUCTION

The detection of high concentrations of glucose in complicated mixtures has attracted an abundance of attention in the food, beverage, and fermentation industries, as well as in the treatment of disease in domestic animals, where glucose mostly acts as a vital substance and is present in high concentrations, from 1 to 10 g/dL [1-8]. Schemes employing highly efficient liquid chromatography and glucose oxidase have been used to predict high glucose concentrations in a complex situation [2-8]. However, most previous approaches were susceptible to several critical disadvantages, including sample destruction, short enzyme lifetime, long measurement time caused by either the catalytic reaction or substance separation process, and high cost resulting from immobilization of the enzyme. To mitigate these issues, optical detection schemes have been adopted for their salient features, including an enzyme-free process, fast response, and the potential for noninvasive detection. Absorption spectroscopy based on

Beer's Law assumes a linear relationship between glucose concentration and absorbance [9-14], which is only good for concentrations below hundreds of mg/dL [15]. It has been noted that a bulky, expensive spectrometer is required to continuously monitor the absorption spectrum [10, 12].

In this paper, an enzyme-free optical method is proposed and demonstrated to predict high concentrations of glucose in a glucose-lactose mixture, by drawing upon the reflective optical power observed at two probe wavelengths. It is noted that lactose in particular is selected as the interfering material to better mimic practical situations, considering that it is one of the most common components of food and beverages [16]. By investigating the wavelength-dependent correlation coefficient ( $CORR(\lambda)$ ) for the reflection spectrum and concentration of pure glucose/lactose solution, conspicuous candidates for the probe wavelengths are initially selected to exhibit a linear relationship between reflective power and glucose/lactose concentration. A set of proportionality coefficients is subsequently derived to develop a predictive

\*Corresponding author: [slee@kw.ac.kr](mailto:slee@kw.ac.kr)

Color versions of one or more of the figures in this paper are available online.

equation, which is eventually validated by comparing predicted and actual glucose concentrations.

## II. PROPOSED GLUCOSE DETECTION USING REFLECTIVE OPTICAL POWER FROM A GLUCOSE-LACTOSE MIXTURE

Rather than using absorption spectroscopy based on Beer's Law, we aim to predict glucose concentrations as high as several g/dL in a glucose-lactose mixture by exploiting the reflective optical power available from the mixture. This power is observed by employing a mirror, without resorting to a spectrometer, which is potentially desirable for noninvasive glucose detection. Actually, the prediction relies on the relative optical power, which is tantamount to the observed optical power normalized with respect to the signal available from pure water. The proposed detection scheme involves the following two equations, implying a linear relationship between the concentration of pure glucose/lactose solution and the corresponding reflective power [13, 17]:

$$\begin{aligned} P_1 &= P_{G1} + P_{L1} = a_1 C_G + b_1 C_L \text{ and} \\ P_2 &= P_{G2} + P_{L2} = a_2 C_G + b_2 C_L \end{aligned} \quad (1)$$

For  $i = 1$  or  $2$ ,  $P_{Gi}$ ,  $P_{Li}$ , and  $P_i$  respectively represent the reflective power for a pure glucose solution, a pure lactose solution, and a glucose-lactose mixture, which are obtained at a probe wavelength  $\lambda_i$ .  $C_G$  and  $C_L$  refer to the concentration of the pure glucose and lactose solutions respectively. The coefficients of proportionality are accordingly designated by  $a_i$  and  $b_i$ .

We have chosen probe wavelengths  $\lambda_1$  and  $\lambda_2$  under the condition that the magnitude of  $CORR(\lambda)$  between the reflection spectra and concentrations is in the vicinity of 1 for both pure glucose and lactose solutions. The reflection spectra for the pure glucose and lactose solution have been normalized relative to the spectrum of pure water. The correlation coefficient is determined by the following equation:

$$CORR(\lambda) = \frac{\sum_{k=1}^n (C_k(\lambda) - \bar{C}(\lambda))(R_k(\lambda) - \bar{R}(\lambda))}{\sqrt{\sum_{k=1}^n (C_k(\lambda) - \bar{C}(\lambda))^2 \sum_{k=1}^n (R_k(\lambda) - \bar{R}(\lambda))^2}} \quad (2)$$

For a total of  $n$  different glucose solutions,  $C_k(\lambda)$  and  $R_k(\lambda)$  respectively denote analyte concentration and reflection spectrum associated with the  $k^{\text{th}}$  glucose solution.  $\bar{C}(\lambda)$  and  $\bar{R}(\lambda)$  are the relevant average concentration and reflection spectrum [18]. This procedure has been similarly applied to the case of lactose as analyte.  $CORR(\lambda)$  may be either positive or negative; a negative value indicates that the corresponding reflection spectrum decreases when the concentration of glucose or lactose increases. When the two probe wavelengths had been determined, the linear coefficients

$a_1$ ,  $b_1$ ,  $a_2$ , and  $b_2$  for glucose and lactose, per Eq. (1), were found by examining the reflective power from a group of reference solutions of glucose and lactose with different concentrations. According to Eq. (1), the glucose concentration is given by  $C_G = (b_2 P_1 - b_1 P_2) / (a_1 b_2 - a_2 b_1)$ . The predictive performance is evaluated in terms of the standard error of prediction ( $SEP$ ):

$$\begin{aligned} SEP &= \sqrt{\frac{1}{n-1} \sum_{k=1}^n (\hat{C}_{Gk} - C_{Gk} - Bias)^2}, \\ \text{for } Bias &= \frac{1}{n} \sum_{k=1}^n (\hat{C}_{Gk} - C_{Gk}) \end{aligned} \quad (3)$$

For a total of  $n$  glucose-lactose mixtures,  $C_{Gk}$  and  $\hat{C}_{Gk}$  are the  $k^{\text{th}}$  actual and predicted glucose concentrations, respectively, while the average difference between the predicted and actual glucose concentrations is expressed by Bias [19]. The  $SEP$ , having the same units of g/dL as the glucose concentration, basically indicates the standard deviation of the error between predicted and actual glucose concentrations.

## III. VALIDATION OF THE PROPOSED GLUCOSE DETECTION SCHEME

To assess our approach for predicting high glucose concentrations in a glucose-lactose mixture, we selected two prominent probe wavelengths that led to high correlations between concentration and reflective optical power. A set of pure glucose and lactose standard solutions of various concentrations was prepared in a volumetric flask by dissolving D-(+)-glucose and  $\alpha$ -lactose powder (Sigma-Aldrich) in deionized (DI) water at room temperature. The standard solution was then poured in a quartz cuvette of path length 10 mm. The glucose concentration was varied from 1 to 45 g/dL, while the lactose concentration was limited to a range of 1 to 9 g/dL, due to its lower solubility. To investigate the reflection spectra for the reference glucose and lactose solutions, we built a test setup as shown in Fig. 1, incorporating a spectrometer (NIRQuest512-2.5, Ocean Optics) and a halogen lamp (HL-2000-FHSA, Ocean Optics), with no bandpass filter inserted. Light from the lamp, which is delivered via the six surrounding fibers belonging to its reflection probe, was shone onto the prepared solution. Light transmitted through the solution was reflected back from a mirror to the central fiber of the reflection probe, for analysis by the spectrometer. During measurement the solutions and detector were entirely isolated from ambient light. The calculated reflection spectra for the two types of solutions are presented in Fig. 2, in the near-infrared regime from  $\lambda = 1000$  to 1700 nm. The resulting  $CORR(\lambda)$  for the glucose and lactose solutions was estimated using Eq. (2). As plotted in Fig. 3, the glucose  $CORR(\lambda)$

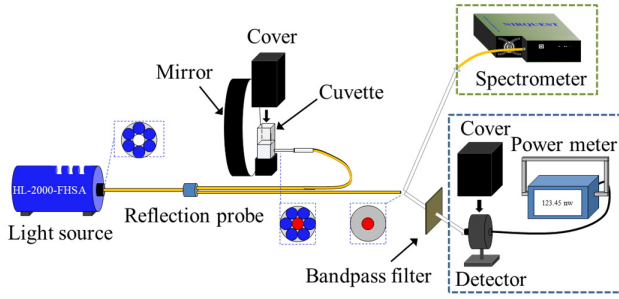


FIG. 1. Test setup for acquiring reflection spectra and monitoring the reflective optical power from the prepared solutions, with a bandpass filter centered at the selected wavelengths.

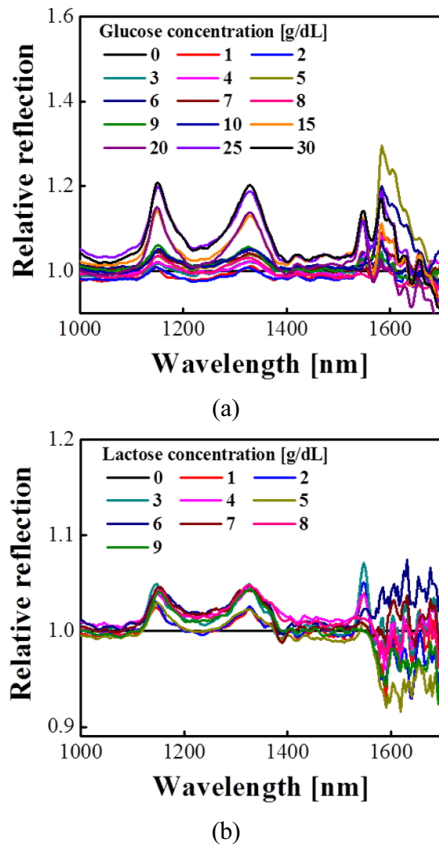


FIG. 2. Measured relative reflection spectra of pure (a) glucose and (b) lactose solutions of various concentrations.

was revealed to be as high as 0.95 over a broad span of wavelengths from  $\lambda = 1135$  to  $1370$  nm. In the case of lactose, the two highest values of  $CORR(\lambda)$  were obtained at  $\lambda = 1160$  and  $1300$  nm. It is noted that lactose yielded a lower  $CORR(\lambda)$  than glucose. In an aqueous solution, lactose exists in two isomeric forms,  $\alpha$ - and  $\beta$ -lactose, with different properties, including specific rotation and solubility. By the process of mutarotation, lactose switches between the two forms to reach an equilibrium state [20]. It is also known that the dissolution of lactose declines with increasing

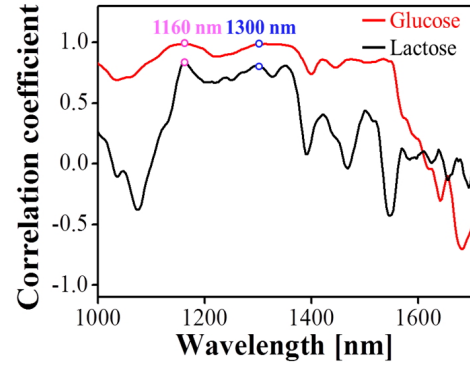


FIG. 3. Calculated correlation coefficients for reflection spectra and concentrations of pure glucose/lactose solutions, in the spectral range from 1000 to 1700 nm.

concentration [21], thus degrading the uniformity of the solution. As a result, the correlation between reflective optical power and lactose concentration weakens with increasing concentration, compared to the case of glucose. As a consequence, the probe wavelengths used for concentration prediction were chosen to be  $\lambda_1 = 1160$  nm and  $\lambda_2 = 1300$  nm, offering substantially high  $CORR(\lambda)$  for both glucose and lactose.

The linear coefficients  $a_1$ ,  $b_1$ ,  $a_2$ , and  $b_2$  of Eq. (1) were determined by analyzing the reflective optical power for the glucose and lactose solutions at the chosen probe wavelengths. As shown in Fig. 1, a photodetector (818-IR, S/N 7736) in combination with an optical power meter (Model-1830-C, Newport) was used to record the reflective power. Two bandpass filters, centered at  $\lambda = 1160$  and  $1300$  nm respectively, were placed between the solution and fiber. As shown in Fig. 4, the measured optical powers  $P_{G1}$ ,  $P_{L1}$ ,  $P_{G2}$ , and  $P_{L2}$  for the glucose and lactose solutions observed at  $\lambda_1 = 1160$  nm and  $\lambda_2 = 1300$  nm increase in apparently linear fashion with analyte concentration. The linear coefficients were accordingly found to be  $a_1 = 5.3625 \times 10^{-3}$ ,  $b_1 = 8.3533 \times 10^{-3}$ ,  $a_2 = 10.0786 \times 10^{-3}$ , and  $b_2 = 14.8262 \times 10^{-3}$ , which may be efficiently utilized to estimate the glucose concentration in a glucose-lactose mixture. Regarding Eq. (1), the final predictive equation is given by  $C_G = (1.7835P_2 - 3.1656P_1) \times 10^3$ , which was employed to predict the glucose concentration in the glucose-lactose mixture by monitoring the optical power. Three pairs of mixtures were prepared, in which glucose and lactose had respective concentrations of 2 and 6 g/dL, 4 and 3 g/dL, and 7 and 3 g/dL. Based on the reflective powers for the mixtures at the predetermined probe wavelengths, we attempted to predict the glucose concentration using the aforementioned predictive equations. For various glucose-lactose mixtures, the comparison between actual and predicted glucose concentrations is shown in Fig. 5. It was finally confirmed that the predicted glucose concentration closely mimics the ideal case, providing an acceptable standard error of prediction  $SEP = 1.17$  g/dL.

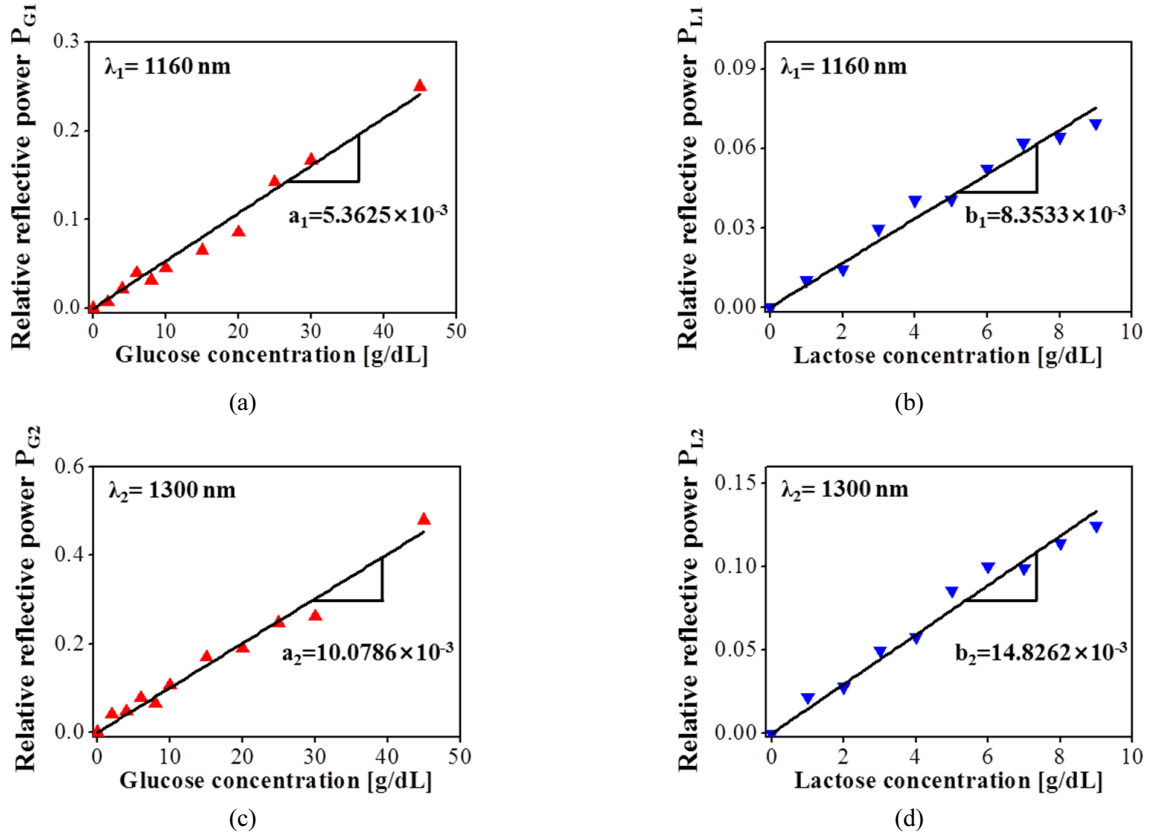


FIG. 4. Fitted linear relationships between concentrations of pure glucose/lactose and the relative reflective power at (a)  $\lambda_1 = 1160$  and (b)  $\lambda_2 = 1300$  nm, where  $a_1$ ,  $b_1$ ,  $a_2$ , and  $b_2$  represent the corresponding linear coefficients.

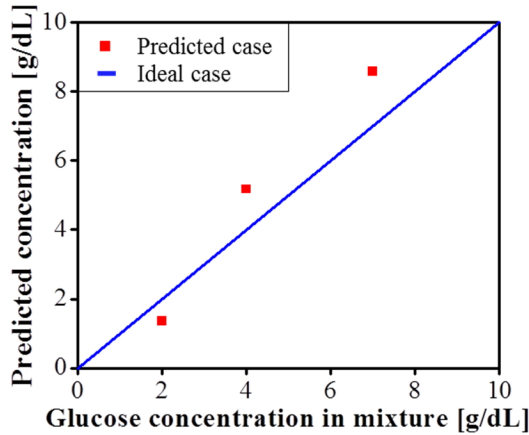


FIG. 5. Comparison of predicted and actual concentrations of glucose in glucose-lactose mixtures.

#### IV. CONCLUSION

An enzyme-free optical scheme for detecting high concentrations of glucose in glucose-lactose mixtures was realized by utilizing the reflective optical power available from the mixture, observed at dual probe wavelengths of 1160 and 1300 nm. The predictive equation was established based on the linear relationship between reflective optical

power and glucose/lactose concentration at the specified wavelengths. For a glucose-lactose mixture, glucose concentrations from 2 to 7 g/dL were successfully estimated, achieving a standard error of prediction of 1.17 g/dL. It is noted that to practically apply the proposed method, for initial calibration a reference solution of pure DI water should be measured.

#### ACKNOWLEDGMENT

This work was supported by a National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIP) (No. 2013-008672), and a research grant from Kwangwoon University in 2015. The authors are grateful to Mr. Vivek R. Shrestha of Kwangwoon University for his helpful discussions, and to Mr. Chanwoo J. Lee of Seoul Foreign School, Seoul, S. Korea, for his contributions to the preparation of the manuscript.

#### REFERENCES

1. T. Sato, K. Katayama, T. Arai, T. Sako, and H. Tazaki, "Simultaneous determination of serum mannose and glucose concentrations in dog serum using high performance liquid

- chromatography," *Res. Vet. Sci.* **84**, 26-29 (2008).
2. G. P. Parpinello and A. Versari, "A simple high-performance liquid chromatography method for the analysis of glucose, glycerol, and methanol in a bioprocess," *J. Chromatogr. Sci.* **38**, 259-261 (2000).
  3. J. P. Yuan and F. Chen, "Simultaneous separation and determination of sugars, ascorbic acid and furanic compounds by HPLC-dual detection," *Food Chem.* **64**, 423-427 (1999).
  4. L. Setti, A. Fraleoni-Morgera, B. Ballarin, A. Filippini, D. Frascaro, and C. Piana, "An amperometric glucose biosensor prototype fabricated by thermal inkjet printing," *Biosens. Bioelectron.* **20**, 2019-2026 (2005).
  5. M. D. Gouda, M. A. Kumar, M. S. Thakur, and N. G. Karanth, "Enhancement of operational stability of an enzyme biosensor for glucose and sucrose using protein based stabilizing agents," *Biosens. Bioelectron.* **17**, 503-507 (2002).
  6. A. M. G. Vasilarou and C. A. Georgiou, "Enzymatic spectrophotometric reaction rate determination of glucose in fruit drinks and carbonated beverages. An analytical chemistry laboratory experiment for food science-oriented students," *J. Chem. Educ.* **77**, 1327-1329 (2000).
  7. R. H. Matthews, P. R. Pehrsson, and M. Farhat-Sabet, *Sugar Content of Selected Foods: Individual and Total Sugars* (U.S. Department of Agriculture, Human Nutrition Information Service, USA, 1987).
  8. B. A. A. Dremel, B. P. H. Schaffar, and R. D. Schmid, "Determination of glucose in wine and fruit juice based on fibre-optic glucose biosensor and flow-injection analysis," *Anal. Chim. Acta.* **225**, 293-301 (1989).
  9. W. B. Martin, S. Mirov, and R. Venugopalan, "Using two discrete frequencies within the middle infrared to quantitatively determine glucose in serum," *J. Biomed. Opt.* **7**, 613-617 (2002).
  10. M. Brandstetter, A. Genner, K. Anic, and B. Lendl, "Tunable external cavity quantum cascade laser for the simultaneous determination of glucose and lactate in aqueous phase," *Analyst* **135**, 3260-3265 (2010).
  11. M. Meinke, G. Muller, H. Albrecht, C. Antoniou, H. Richter, and J. Lademann, "Two-wavelength carbon dioxide laser application for in-vitro blood glucose measurements," *J. Biomed. Opt.* **13**, 014021 (2008).
  12. S. Yu, D. Li, H. Chong, C. Sun, H. Yu, and K. Xu, "In vitro glucose measurement using tunable mid-infrared laser spectroscopy combined with fiber-optic sensor," *Biomed. Opt. Express* **5**, 275-286 (2014).
  13. H. Fisher, R. G. Hansen, and H. W. Norton, "Quantitative determination of glucose and galactose," *Anal. Chem.* **27**, 857-859 (1955).
  14. Y. Mendelson, A. C. Clermont, R. A. Peura, and B. C. Lin, "Blood glucose measurement by multiple attenuated total reflection and infrared absorption spectroscopy," *IEEE Trans. Biomed. Eng.* **37**, 458-465 (1990).
  15. S. Nielsen, *Food Analysis* (Springer Science & Business Media, USA, 2010).
  16. T. Katsu, X. Zhang, and G. A. Rechnitz, "Simultaneous determination of lactose and glucose in milk using two working enzyme electrodes," *Talanta* **41**, 843-848 (1994).
  17. J. H. Rodriguez-Rodriguez, F. Martinez-Pinon, J. A. Alvarez-Chavez, D. Jaramillo-Vigueras, and E. G. Robles-Pimentel, "Direct optical techniques for the measurement of water content in oil-paper insulation in power transformers," *Meas. Sci. Technol.* **22**, 065706 (2011).
  18. R. M. Vogel, "The probability plot correlation coefficient test for the normal, lognormal, and gumbel distributional hypotheses," *Water Resour. Res.* **22**, 587-590 (1986).
  19. K. H. Esbensen, D. Guyot, F. Westad, and L. P. Houmoller, *Multivariate Data Analysis - In Practice: An Introduction to Multivariate Data Analysis and Experimental Design*, 5th ed. (Multivariate Data Analysis, Denmark, 2002).
  20. P. F. Fox, T. Uniacke-Lowe, P. L. H. McSweeney, and J. A. O'Mahony, *Dairy Chemistry and Biochemistry*, 2nd ed. (Springer, 2015).
  21. P. L. H. McSweeney and P. F. Fox, *Advanced Dairy Chemistry: Volume 3: Lactose, Water, Salts and Minor Constituents*, 3rd ed. (Springer Science & Business Media, 2009).