CHAPTER 1

Principles of Hyperspectral Imaging Technology

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1.1. INTRODUCTION

During the past few decades a number of different techniques have been explored as possible instrumental methods for quality evaluation of food products. In recent years, *hyperspectral imaging* technique has been regarded as a smart and promising analytical tool for analyses conducted in research, control, and industries. Hyperspectral imaging is a technique that generates a spatial map of spectral variation, making it a useful tool in many applications. The use of hyperspectral imaging for both automatic target detection and recognizing its analytical composition is relatively new and is an amazing area of research. The main impetus for developing a hyperspectral imaging system was to integrate spectroscopic and imaging techniques to enable direct identification of different components and their spatial distribution in the tested sample. A hyperspectral imaging system produces a two-dimensional spatial array of vectors which represents the spectrum at each pixel location. The resulting three-dimensional dataset containing the two spatial dimensions and one spectral dimension is known as the *datacube* or hypercube (Chen *et al.*, 2002; Kim *et al.*, 2002; Mehl *et al.*, 2004; Schweizer & Moura, 2001). The advantages of hyperspectral imaging over the traditional methods include minimal sample preparation, nondestructive nature, fast acquisition times, and visualizing spatial distribution of numerous chemical compositions simultaneously. The hyperspectral imaging technique is currently tackling many challenges to be accepted as the most preferable analytical tool in identifying compositional fingerprints of food

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products and their authentication. The need for fast and reliable methods of authenticity and object identification has increased the interest in the application of hyperspectral imaging for quality control in the agricultural, pharmaceutical, and food industries. Moreover, enhancement in instrumental developments, the availability of high-speed computers, and the development of appropriate chemometric procedures will allow this technique to be dominant in the future. This chapter presents the fundamentals, characteristics, configuration, terminologies, merits and demerits, limits and potential of hyperspectral imaging. Basics and theoretical aspects relating to this technique, the information that can be supplied, and the main features of the instrumentation are presented and briefly discussed. The final part of the chapter concerns a general overview of the main steps involved in analyzing hyperspectral images. The potential applications of hyperspectral imaging in food analysis will be explained in more detail in the relevant chapters of this book

1.1.1. The Necessity for Automating Quality Assessment

With increased expectations for food products with high quality and safety, the need for accurate, fast, and objective quality determination of these characteristics continues to grow. Quality assurance is one of the most important goals of any industry. The ability to manufacture high-quality products consistently is the basis for success in the highly competitive food industry. It encourages loyalty in customers and results in an expanding market share. The quality assurance methods used in the food industry have traditionally involved human visual inspection. Such methods are tedious, laborious, time-consuming, and inconsistent. As plant throughput increased and quality tolerance tightened, it became necessary to employ automatic methods for quality assurance and quality control (Gunasekaran, 1996). Also, the increased awareness and sophistication of consumers have created the expectation for improved quality food products. Consumers are always demanding superior quality of food products, i.e., higher quality for an individual food item, consistency of products in a batch, and enhanced food safety as a whole (Nagata *et al.*, 2005). This in turn has increased the need for enhanced quality monitoring. In general, automation of a quality assessment operation not only optimizes quality assurance but more importantly it also helps in removing human subjectivity and inconsistency. Moreover, automation usually increases the productivity and changes the character of the work, making it less arduous and more attractive. Considering the fact that the productivity of a person working in a mechanized and automated environments is approximately ten times that of a manual worker, this has

stimulated progress in the development of many novel sensors and instruments for the food industry, often by technology transfer from other industrial sectors, including medical, electronic, and nonclinical sectors (Abdullah *et al.*, 2004). If quality evaluation is achieved automatically, production speed and efficiency can be improved drastically in addition to increased evaluation accuracy, with an accompanying reduction in production costs.

1.2. RELATIONSHIP BETWEEN SPECTROSCOPY, IMAGING, AND HYPERSPECTRAL IMAGING

In the past two decades, considerable progress has been accomplished in the development of new sensing technologies for quality and safety inspection of agricultural and food products. These new sensing technologies have provided us with unprecedented capabilities to measure, inspect, sort, and grade food products effectively and efficiently. Consequently, some smart methods to evaluate quality and quality-related attributes have been developed using advanced techniques and instrumentation. Most recently, the emphasis has been on developing sensors for real-time, nondestructive systems. As a result, automated visual inspection by computer-based systems has been developed in the food industry to replace the traditional inspection by human inspectors because of its cost-effectiveness, consistency, superior speed, and accuracy. *Computer vision* technology utilizing image processing routines is one alternative which became an integral part of the industry's move towards automation. Combined with an illumination system, a computer vision system is typically based on a personal computer in connection with electrical and mechanical devices to replace human manipulative effort in the performance of a given process (Du & Sun, 2006). Image processing and image analysis are the core of computer vision, involving mathematics, computer science and software programming. This system has a great ability in evaluation cycle to apply the principle: several objects per second instead of several seconds per object.

Unfortunately, the computer vision system has some drawbacks that make it unsuitable for certain industrial applications. It is inefficient in the case of objects of similar colours, inefficient in the case of complex classifications, unable to predict quality attributes (e.g. chemical composition), and it is inefficient for detecting invisible defects. Since machine vision is operated at visible wavelengths, it can only produce an image registering the external view of the object and not its internal view. Situations exist whereby

food technologists need to look inside the object in a noninvasive and nondestructive manner. For instance, food technologists need to measure and map the water content of food in order to assess its microbiological stability and to implement risk analysis as defined by the hazard analysis critical control point (HACCP) (Abdullah *et al.*, 2004). Therefore, external attributes such as size, shape, colour, surface texture, and external defects can easily be evaluated by ordinary means (e.g. RGB colour camera). However, internal structures are difficult to detect with relatively simple and traditional imaging means, which cannot provide enough information for detecting internal attributes (Du & Sun, 2004).

Since quality is not a single attribute but comprises many properties or characteristics (Abbott, 1999; Noh & Lu, 2005), measurement of the *optical properties* of food products has been one of the most successful nondestructive techniques for quality assessment to provide several quality details simultaneously. Optical properties are based on reflectance, transmittance, absorbance, or scatter of polychromatic or monochromatic radiation in the ultraviolet (UV), visible (VIS), and near-infrared (NIR) regions of the electromagnetic spectrum which can be measured by spectral instruments. A quality index for the product can be based on the correlation between the spectral response and a specific quality attribute of the product, usually a chemical constituent (Park *et al.*, 2002). Diffusely reflected light contains information about the absorbers near the surface of a material. Recently, optical techniques using near-infrared spectroscopy (NIRS) have received considerable attention as a means for nondestructive sensing of food quality. NIRS is rapid, nondestructive, and relatively easy to implement for on-line and off-line applications. More importantly, NIRS has the potential for simultaneously measuring multiple quality attributes. In these spectroscopic techniques, it is possible to obtain information about the sample components based on the light absorption of the sample, but it is not easy to know the position/location information. On the other hand, it is easy to know the position of certain features by naked eye or computer vision systems, but it is not easy to conduct the quantitative analysis of a component. The combination of the strong and weak points of visible/near-infrared spectroscopic techniques and vision techniques is the *hyperspectral imaging* technique, which is also called *imaging spectroscopy* or *imaging spectrometry*, even though the meaning is different (spectrometry—"measuring", spectroscopy—"seeing", hyperspectral—"many bands"). Because hyperspectral imaging techniques overcome the limits of spectroscopic techniques and vision techniques, they have emerged as a powerful technique in agricultural and food systems. Based on hyperspectral imaging techniques,

multispectral imaging system can be built for real-time implementations (Lee *et al.*, 2005).

While a grayscale image typically reflects the light intensity over the electromagnetic spectrum in a single band, a colour image reflects the intensity over the red, green, and blue bands of the spectrum. Increasing the number of bands can greatly increase the amount of information from an image. Hyperspectral images commonly contain information from several bands with different resolution values. Hyperspectral imaging has been invented to integrate spectroscopic and spatial (imaging) information which otherwise cannot be achieved with either conventional imaging or spectroscopic techniques. It involves measuring the intensity of diffusely reflected light from a surface at one or more wavelengths with relatively narrow bandpasses. Hyperspectral imaging goes beyond conventional imaging and spectroscopy to acquire both spectral and spatial information from an object simultaneously. *Imaging* technique is essentially the science of acquiring spatial and temporal data information from objects using a digital camera, whereas *spectroscopy* is the science of acquiring and explaining the spectral characteristics of an object to describe light intensities emerging from its molecules at different wavelengths and thus provide a precise fingerprint of that object. Since image data are considered two-dimensional, by adding a new dimension of "spectrum" information, the hyperspectral image data can be perceived as a three-dimensional datacube (Chao *et al.*, 2001). Hyperspectral imaging, like other spectroscopy techniques, can be carried out in reflectance, transmission or fluorescence modes. While the majority of published research on hyperspectral imaging has been performed in reflectance mode, transmission and emission modes have also been investigated. In brief, the main differences and advantages of hyperspectral imaging over conventional imaging and spectroscopic techniques are outlined in Table 1.1.

Table 1.1 Main differences among imaging, spectroscopy, and hyperspectral imaging techniques

Features	Imaging	Spectroscopy	Hyperspectral imaging
Spatial information	√	×	√
Spectral information	×	√	√
Multi-constituent information	×	√	√
Building chemical images	×	×	√
Flexibility of spectral information extraction	×	×	√

1.2.1. Advantages of Hyperspectral Imaging

The rich information content and outstanding feature identification capabilities of hyperspectral imaging make it highly suitable for numerous applications. However, the technology also has some demerits that need to be considered before its implementation in food quality assessment regimes. These are covered in the following section. The foremost advantages of using hyperspectral imaging technology in food analysis can be summarized in the following points:

No sample preparation is required.

It is a chemical-free assessment method, which enables safety and environmental protection by thoroughly eliminating pollutant solvents, chemicals and/or potentially dangerous reagents during analyses.

Once the calibration model is built and validated, it becomes an extremely simple and expeditious analysis method.

It is a noninvasive, and nondestructive method, so that the same sample could be used for other purposes and analyses.

It is eventually economic compared with traditional methods, owing to the savings in labor, time, and reagent cost in addition to the large saving in the cost of waste treatments.

Rather than collecting a single spectrum at one spot on a sample, as in spectroscopy, hyperspectral imaging records a spectral volume that contains a complete spectrum for every spot (pixels) in the sample.

It has the flexibility in choosing any region of interest (ROI) in the image even after image acquisition. Also, when an object or a ROI in the object presents very obvious spectral characteristics, that region could be selected and its spectrum is saved in a spectral library.

Due to its high spectral resolution, hyperspectral imaging provides both qualitative and quantitative measurements.

It is able to determine several constituents simultaneously in the same sample.

One of the strategic advantages of hyperspectral imaging is that it allows for the visualization of different biochemical constituents presented in a sample based on their spectral signatures because regions of similar spectral properties should have similar chemical composition. This

process is called building *chemical images*, or chemical mapping, for constructing detailed maps of the surface composition of foods which traditionally requires use of intense laboratory methods. This approach will be explained in more detail in Chapter 6.

The greater spectral information residing in the spectral images allows many different objects to be detected and distinguished even if they have similar colors, morphological features or overlapping spectra.

The spatial distribution and concentration of the chemical composition in the product can be obtained, not just the bulk composition.

Its ability to build chemical images permits labeling of different entities in a sample simultaneously and quantitative analysis of each entity. Therefore, it enables documentation of the chemical composition of the product. Such documentation allows different pricing and labeling to be used in sorting food products with different chemical compositions according to market requirements, consumer preference, and/or product specifications.

If the high dimensionality of hyperspectral imaging were reduced to form multispectral imaging by choosing some optimal wavelengths for certain classifications, the technology would be incomparable for process monitoring and real-time inspection.

1.2.2. Disadvantages and Constraints of Hyperspectral Imaging

In spite of the aforementioned advantages, hyperspectral imaging does have some disadvantages, which can be summarized as follows:

Hyperspectral images contain a substantial amount of data, including much redundant information, and pose considerable computational challenges.

It takes a long time for image acquisition and analysis, therefore hyperspectral imaging technology has to a very limited extent been directly implemented in on-line systems for automated quality evaluation purposes.

From an analyst's point of view, one of the main analytical drawbacks of hyperspectral imaging technique is that it is an indirect method, which means that it needs standardized calibration and model transfer procedures.

Similar to all spectroscopic techniques, spectral data extracted from any location of the image contain a series of successive overlapping bands, which are difficult to assign to specific chemical groups.

One major factor that limits its industrial applications for food inspection is the hardware speed needed for rapid image acquisition and analysis of the huge amount of data collected.

Hyperspectral data suffer from the well-known problem of multicollinearity; although some multivariate analysis techniques like principal component regression (PCR) and partial least square (PLS) are often employed to overcome this problem. However, the effects of multicollinearity in data can only be reduced but not completely removed by PCR and PLS. In this aspect, variable selection is advantageous in the sense that not only can it improve the predictive power of the calibration model, but also it can simplify the model by avoiding repetition of information or redundancies and irrelevant variables.

Hyperspectral imaging is not suitable in some cases, such as liquids or homogenous samples, because the value of imaging lies in the ability to resolve spatial heterogeneities in samples. Imaging a liquid or even a suspension has limited use as constant sample motion serves to average spatial information, unless ultra-fast recording techniques are employed as in fluorescence correlation microspectroscopy or fluorescence lifetime imaging microscopy (FLIM) observations where a single molecule may be monitored at extremely high detection speed. Similarly, there is no benefit in imaging a truly homogeneous sample, as a single point spectrometer will generate the same spectral information. Of course the definition of homogeneity is dependent on the spatial resolution of the imaging system employed.

To identify and detect different objects unambiguously in the same image, these objects must exhibit characteristic absorption features. Furthermore, if an object has diagnostic absorption features, it must be present at a minimum concentration or converge in a pixel to be detected.

Depending on the spatial resolution and the structure of the sample investigated, spectra from individual image pixels may not represent a pure spectrum of one singular material, but a mixed spectrum consisting of spectral responses of the various materials that cover the region of interest (ROI) selected from the sample.

In a hyperspectral imaging system it is time-consuming to acquire the spectral and spatial information of the entire sample, and therefore it is not

practical to implement such a system on-line as it is. However, by means of analyzing the hyperspectral imaging data, it is possible to select a few effective and suitable wavebands for building a multispectral imaging system to meet the speed requirement of production lines (Xing *et al.*, 2006). The problem caused by the huge amount of data generated in hyperspectral imaging can be overcome by using data reduction schemes in such a way that only those wavelengths and spatial locations of special interest are selected. In this way, the amount of data can be effectively reduced, which will benefit later data processing. Therefore the hyperspectral imaging experiment is usually conducted off-line in the laboratory to select some optimal wavelengths for later multispectral imaging measurements suitable for on-line applications (Chao *et al.*, 2002; Mehl *et al.*, 2004). Once the optimal inspection bands are identified, an automatic inspection system using only these bands can be designed and then industrially implemented. Such a method has been increasingly used with computers becoming faster and more powerful, and it has now entered a new era of industrial applications for on-line evaluation of food and agricultural products. Nowadays, a significant number of scientific articles are published annually on hyperspectral and multispectral imaging for various applications. Moreover, several manufacturers specialized in spectral systems have emerged in the market to sell not only the spectral components but also the whole hyperspectral imaging units.

1.3. FUNDAMENTALS OF HYPERSPECTRAL IMAGING

In order to use the hyperspectral imaging technology, a good understanding of the theory behind the technique is required. Therefore, some basic information about spectroscopy will be provided in this section. The electromagnetic spectrum and the nature of light and its properties are also described to allow the reader to gain knowledge about the importance of light in hyperspectral imaging. Furthermore, definitions of basic terms, such as wavelength, waveband, frequency, spectral signature, and spectrum, are briefly given. Detailed descriptions can be found in many optics and physics textbooks (e.g. Hecht, 2002).

1.3.1. Basics of Spectroscopy

The root of *spectrometric technique* dates back to 1665, when Sir Isaac Newton described the concept of dispersion of light and the optomechanical hardware of a spectrometer after he passed light through a *prism* and observed the splitting of light into colors. In particular, visible and near-infrared

spectroscopy is an established technique for determining chemical constituents in food products. These instruments use gratings to separate the individual frequencies of the radiation leaving the sample. The development of an NIR spectrometric technique for assessing quality traits in food products relies on the collection of spectra of the produce and developing a calibration equation to relate this spectral data to the quality trait ascertained using a standard laboratory method. In NIR quantitative analysis, this is typically called a calibration equation. The difference between failing and succeeding in this task is greatly dependent on the quality of the reference values associated with the samples in the calibration set. Nevertheless, once this learning stage is concluded, the final result is perhaps close to the result of an ideal analytical method (Pieris *et al.*, 1999).

Basically, spectroscopic methods provide detailed fingerprints of the biological sample to be analysed using physical characteristics of the interaction between electromagnetic radiation and the sample material, such as reflectance, transmittance, absorbance, phosphorescence, fluorescence, and radioactive decay. *Spectroscopic analysis* exploits the interaction of electromagnetic radiation with atoms and molecules to provide qualitative and quantitative chemical and physical information contained within the wavelength spectrum that is either absorbed or emitted. Among these spectroscopic techniques, NIR spectroscopy is one of the most successful within the food industry. The absorption bands seen in this spectral range arise from overtones and combination bands of O–H, N–H, C–H, and S–H stretching and bending vibrations that enable qualitative and quantitative assessment of chemical and physical features. Therefore, NIR could be applied to all organic compounds rich in O–H bonds (such as moisture, carbohydrate and fat), C–H bonds (such as organic compounds and petroleum derivatives), and N–H bonds (such as proteins and amino acids). In a given wavelength range, some frequencies will be absorbed, others (that do not match any of the energy differences between vibration response energy levels for that molecule) will not be absorbed, while some will be partially absorbed. This complex relation between the intensity of absorption and wavelength constitutes the absorption spectra of a substance or sample (Pasquini, 2003). Since all biological substances contain thousands of C–H, O–H, and N–H molecular bonds, the exposure of a sample to NIR radiation results in a complex *spectrum* that contains qualitative and quantitative information about the physical and chemical computational changes of that sample.

Indeed, modern NIR spectroscopy technique requires a low-noise spectrometer, computerized control of the spectrometer and data acquisition, and the use of multivariate mathematical and statistical computer algorithms to

analyse the data. The bonds of organic molecules change their vibration response energy when irradiated by NIR frequencies and exhibit absorption peaks through the spectrum. Thus, qualitative and quantitative chemical and physical information is contained within the wavelength spectrum of absorbed energy (Carlomagno *et al.*, 2004). However, NIR spectroscopic techniques rely on measuring only the aggregate amount of light reflected or transmitted from a specific area of a sample (point measurement where the sensor is located), and do not give information on the spatial distribution of light in the sample. Besides, when the samples are presented to the spectrometers, their homogeneity is an important issue, since a traditional spectrometer integrates the spatial information present, e.g. in a cuvette. This fact does not influence the measurements when the sample is in the liquid or gaseous phase, but in the case of a solid sample (like all agro-food products), this means losing a great deal of information since there are many cases in which the mapping of some characteristic property spectrally identifiable is of the utmost importance. This greatly limits the ability of NIR spectroscopy to quantify structurally related properties and spatial-related distribution. The logical solution would be the use of hyperspectral imaging, but such a technique imposes major technological challenges both from the hardware and software point of view that should be carefully evaluated before starting any research project.

1.3.2. Importance of Light in Hyperspectral Imaging

In modern physics, the discipline of studying light and interaction of light with matter is called optics. Yet while light enables us to see, we cannot see light itself. In fact, what we see depends fundamentally on the properties of light as well as the physical and physiological processes of our interpretation of the scenes. By the end of the nineteenth century, it seemed that the question of the nature of light had been conclusively settled. *Light* is a nonmaterial wave composed of oscillating electric and magnetic fields and, being nonmaterial, the wave can travel through a vacuum without the aid of a material substance (medium). Through the development of quantum theory during the twentieth century, it has been proved by several investigations that under certain circumstances light behaves as a wave, while under different circumstances it behaves as a stream of massless particles. Thus, light has a dual nature. It displays a wave nature in some experiments and particle-like behavior in others. Therefore, it was also neatly and precisely assumed that light consists of a stream of particles, called *photons*, that travel at the speed of light and carry an amount of energy proportional to the light frequency. Depending on the circumstances, when light behaves as

a wave it is characterized by a speed, wavelength, and frequency; when considered as particles, each particle has an energy related to the frequency of the wave, given by the following *Planck's relation*:

$$E = hf \tag{1.1}$$

where E is the energy of the photon, h is Planck's constant (6.626×10^{-34} J.s), and f is the frequency. When light interacts with a single atom and molecule, its behavior depends on the amount of energy per quantum it carries.

During the nineteenth century there was an explosive increase in our understanding of the properties of light and its behaviors. Wave interference and polarization were discovered, and the speed of light was measured in different media. Instruments using prism and diffraction gratings gave rise to analysis of light spectra from various sources and the field of spectroscopy was born. These spectra became the key to understanding the structure of the atom and discovering numerous characteristics of molecules. In hyperspectral imaging, *light* plays a crucial role in the system in order to see clearer, farther, and deeper and to gain detailed information about different objects under investigation. A hyperspectral imaging system can capture light from frequencies beyond the visible light range. This can allow extraction of additional information that the human eye fails to capture.

1.3.3. Electromagnetic Spectrum

Electromagnetic radiation is a unique phenomenon that takes the form of self-propagating waves in a vacuum or in matter. It consists of electric and magnetic field components that oscillate in phase perpendicular to each other and perpendicular to the direction of energy propagation. The *electromagnetic spectrum*, as shown in Figure 1.1, consists of several categories (or regions), including gamma rays, X-rays, ultraviolet radiation (UV), visible light (VIS), infrared radiation (IR)—divided into near-infrared (NIR), mid-infrared (MIR), and far-infrared (FIR) regions—microwaves and radio waves (FM and AM). Each region corresponds to a specific kind of atomic or molecular transition corresponding to different energies. It is important to indicate that wavelength increases to the right and the frequency increases to the left. These categories are classified in the order of increasing wavelength and decreasing frequency. It has been convenient to divide the spectrum into these categories, even though the division is arbitrary and the categories sometimes overlap. The small region of frequencies with an extremely small range of wavelengths between 400 and 700 nm is sensed by the eyes of

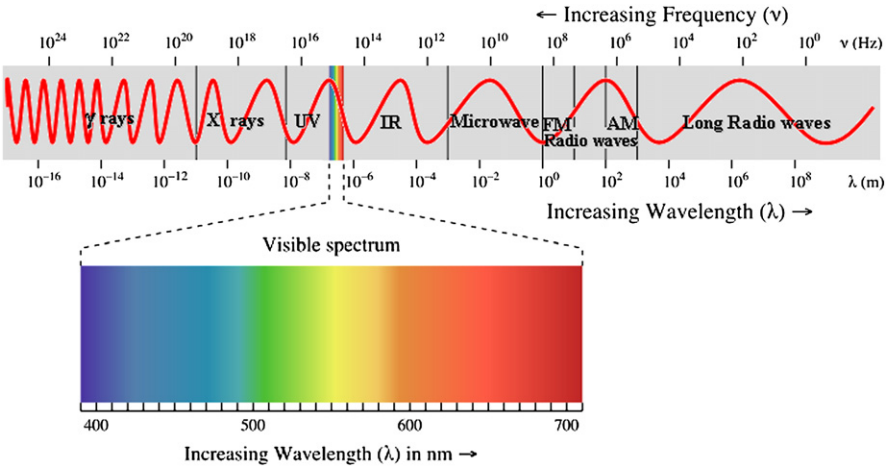

FIGURE 1.1 *Electromagnetic spectrum with visible spectrum (light) magnified. (Full color version available on http://www.elsevierdirect.com/companions/9780123747532/)*

humans and various organisms and is what we call the *visible spectrum*, or *light*.

Light waves are electromagnetic and thus consist of an oscillating electric field perpendicular to and in phase with an oscillating magnetic field. As with all types of waves, the frequency f of an electromagnetic wave is determined by the frequency of the source. The speed of light in a vacuum is defined to be exactly $c = 299{,}792{,}458\ \mathrm{m\,s^{-1}}$ (about 186,282.397 miles per second) which is usually rounded to $3.0 \times 10^8\ \mathrm{m\,s^{-1}}$. In general, an electromagnetic wave consists of successive troughs and crests, and the distance between two adjacent crests or troughs is called the *wavelength*. Waves of the electromagnetic spectrum vary in size, from very long radio waves like the size of a building to very short gamma rays smaller than atom nuclei. *Frequency* (f) is inversely proportional to wavelength (λ), according to the equation of the speed of the wave (υ) which is equal to c in a vacuum:

$$\upsilon = f\lambda \tag{1.2}$$

As waves cross boundaries between different media, their speeds change but their frequencies remain constant. All forms of waves, such as sound waves, water waves, and waves on a string, involve vibrations that need some material to support the wave or media to be conveyed. In the case of electromagnetic waves travelling through empty space, however, no material is needed to support the wave.

1.3.4. Interaction of Light with the Sample

The rationale for the development of a hyperspectral imaging system as a tool for nondestructive food analysis is based on the physical understanding of the interaction of light photons with the molecular structure of food samples. Indeed, studying the subject of interaction of light with biological materials and food samples is of paramount importance in identifying molecules based on their intrinsic properties in order to find their functions, to monitor interactions between different molecules, to detect morphological changes within biological materials, and to correlate changes that occur in the samples with the relevant physiological disorders or disease. In fact, all materials, including food samples, continuously emit and absorb energy by lowering or raising their molecular energy levels. The strength and wavelengths of emission and absorption depend on the nature of the material. Basically, when an electromagnetic wave (from an illumination unit) strikes the surface of a sample, the wave may be partly or totally reflected, and any nonreflected part will penetrate into the material. If a wave passes through a material without any attenuation, the material is called transparent. A material with partial attenuation is known as semitransparent, and a material through which none of the incoming radiation penetrates is called opaque. Most gases are rather transparent to radiation, while most solids (like raw food samples) tend to be strong absorbers for most wavelengths, making them opaque over a distance of a few nanometres to a few micrometres.

Visible light reflected, emitted or transmitted from a product carries information used by inspectors and consumers to judge several aspects of its quality. However, human vision is limited to a small region of the spectrum (as shown in Figure 1.1), and some quality features respond to wavelengths in regions outside the visible spectrum. The characteristics of the radiation that leaves the surface of the product depend on the properties of the product and the incident radiation. When radiation from the lighting system illuminates an object, it is transmitted through, reflected or absorbed. These phenomena are referred to as optical properties. Thus, determining such *optical* characteristics of an agricultural product can provide information related to quality factors of the product. When a sample is exposed to light, some of the incident light is reflected at the outer surface, causing specular reflectance (mirror-like reflectance), and the remaining incident energy is transmitted through the surface into the cellular structure of the sample where it is scattered by the small interfaces within the tissue or absorbed by cellular constituents (Birth, 1976). This is called diffuse reflection, where incoming light is reflected in a broad range of directions. Even when a surface exhibits only specular reflection with no diffuse reflection, not all of the light is

necessarily reflected. Some of the light may be absorbed by the materials. Additionally, depending on the type of material behind the surface, some of the light may be transmitted through the surface. For opaque objects such as most food products, there is no transmission. The detected energy is converted by the spectrometers into *spectra*. These spectra are sensitive to the physical and chemical states of individual constituents. The high spectral signal-to-noise ratio obtained from modern instruments means that even constituents present in quite low concentrations can be detected (Gao *et al.*, 2003).

Most light energy penetrates only a very short distance and exits near the point of entry; this is the basis for color. However some penetrates deeper into the tissues and is altered by differential absorbance of various wavelengths before exiting and therefore contains useful chemometric information. Such light may be called diffuse reflectance, body reflectance, diffuse transmittance, body transmittance or interactance (Abbott, 1999). Meanwhile, the interactions of constituents within product cells alter the characteristic absorbance wavelength and cause many overlapping absorbances (Park *et al.*, 2002). In an attempt to determine the light penetration depth in fruit tissue for each wavelength in the range from 500 to 1900 nm, Lammertyn *et al.* (2000) found that the penetration depth in apple fruit is wavelength-dependent: up to 4 mm in the 700–900 nm range and between 2 and 3 mm in the 900–1900 nm range. In addition, the absorbed light can also be re-emitted (fluorescence), usually at longer wavelengths. A number of compounds emit fluorescence in the VIS region of the spectrum when excited with UV radiation; these compounds are called *fluorophores*. Fluorophores are a functional group in a molecule that will absorb energy of a specific wavelength and re-emit energy at a different, specific wavelength. The amount of the emitted energy and the wavelength at which the energy emits depend on both the fluorophore and the chemical environment of the fluorophore. The optical properties and fluorescence emission from the object are integrated functions of the angle and wavelength of the incident light and chemical and physical composition of the object (Chen *et al.*, 2002). Fluorescence refers to the phenomenon that light of short wavelengths is being absorbed by molecules in the sample tissue with subsequent emission of longer wavelength light. The fluorescence technique has been used for investigating biological materials, detecting environmental, chemical, and biological stresses in plants, and monitoring food quality and safety (Noh & Lu, 2005).

On the other hand, absorption and scattering are two basic phenomena as light interacts with biological materials. Light absorption is related to certain chemical constituents in agro-food samples, such as sugar, acid, water, etc. Modern reflectance NIR spectrometers measure an aggregate amount of light

reflected from a sample, from which light absorption may be estimated and then related to certain chemical constituents. However, scattering is a physical phenomenon that is dependent on the density, cell structures, and cellular matrices of fruit tissue. NIR does not provide quantitative information on light scattering in the sample (Lu, 2004; Peng & Lu, 2005). If both absorption and scattering are to be measured, more significant information about the chemical and physical/mechanical properties of food products could be gained (Lu, 2003a).

1.3.5. Terminology

In dealing with a hyperspectral imaging system, some familiarity with technical information, essential expressions, and definitions will be useful. In this section, basic terminologies normally used in hyperspectral imaging will be highlighted and differentiation among them will be discussed.

1.3.5.1. Spectral range

The *spectral range* describes the wavelength regions covered by the hyperspectral imaging system. Spectral imaging instruments could cover either the ultraviolet, visible, near-infrared or infrared wavelengths based on the required application. Hyperspectral imaging system in the visible and very near-infrared range 380–800 nm or 400–1000 nm is the most widely used in food analysis applications. Nowadays, hyperspectral imaging systems in the range 900–1700 nm that provide the accuracy required in today's most challenging applications in food analysis are available. Moreover, some hyperspectral imaging systems that cover the shortwave-infrared (SWIR) region (900–2500 nm) are currently produced by many manufacturers to serve as significant tools in numerous applications in food and agricultural analyses, chemical imaging, and process analytical technologies.

1.3.5.2. Spectral resolution

The *spectral resolution* of the hyperspectral imaging system is related to its spectrograph as a measure of its power to resolve features in the electromagnetic spectrum. Spectral resolution is defined as the absolute limit of the ability of a hyperspectral imaging system to separate two adjacent monochromatic spectral features emitted by a point in the image. Spectral resolution is a measure of the narrowest spectral feature that can be resolved by a hyperspectral imaging system. The magnitude of spectral resolution is determined by the wavelength dispersion of the spectrograph and the sizes of the entrance and exit apertures. The goal of any spectral imaging system

should be to accurately reconstruct the true spectral profile of an emitting light from all points in the tested sample.

1.3.5.3. Spatial resolution

The *spatial resolution* of the hyperspectral imaging system determines the size of the smallest object that can be seen on the surface of the specimen by the sensor as a distinct object separate from its surroundings. *Spatial resolution* also determines the ability of a system to record details of the objects under study. Higher spatial resolution means more image detail explained. In other words, spatial resolution is defined as the area in the scene that is represented by one image pixel. For practical purposes the clarity of the image is decided by its spatial resolution, not the number of pixels in an image. The parameter most commonly used to describe spatial resolution is the *field of view* (FOV). In effect, spatial resolution refers to the number of pixels per unit length. The spatial resolution is determined by the pixel size of the two-dimensional camera and the objective lens as the spectrograph is designed with a unity magnification.

1.3.5.4. Band numbers

The number of bands is one of the main parameters that characterize hyperspectral imaging systems. Based on the type of spectral imaging system, i.e. multispectral or hyperspectral, the number of spectral bands could vary from a few (usually fewer than 10) in multispectral imaging to about 100–250 spectral bands in the electromagnetic spectrum in the case of hyperspectral imaging. However, the band number is not the only and decisive criterion for choosing a hyperspectral system for certain applications; the second important criterion is the bandwidth.

1.3.5.5. Bandwidth

The *bandwidth* is a parameter that is defined as the full width at half maximum (FWHM) response to a spectral line, describing the narrowest spectral feature that can be resolved by spectrography. Bandwidth should not be interchanged with the spectral sampling intervals, indicating that the spectral distance between two contiguous bands is the same without referring to their bandwidth.

1.3.5.6. Signal-to-noise ratio (SNR or S/N)

The *signal-to-noise ratio* (SNR) is the ratio of the radiance measured to the noise created by the detector and instrument electronics. In other words, signal-to-noise ratio compares the level of a desired signal to the level of background noise. In hyperspectral imaging systems, the SNR is always

wavelength-dependent because of overall decreasing radiance towards longer wavelengths. The higher the ratio, the less obtrusive the background noise is.

1.3.5.7. Spectral signature

Hyperspectral imaging exploits the fact that all materials, due to the difference of their chemical composition and inherent physical structure, reflect, scatter, absorb, and/or emit electromagnetic energy in distinctive patterns at specific wavelengths. This characteristic is called *spectral signature* or *spectral fingerprint*, or simply *spectrum*. Every image element (*pixel*) in the hyperspectral image contains its own spectral signature. Briefly, spectral signature is defined as the pattern of reflection, absorbance, transmittance, and/or emitting of electromagnetic energy at specific wavelengths. In principle, the spectral signature can be used to uniquely characterize, identify, and discriminate by class/type any given object(s) in an image over a sufficiently broad wavelength band (Shaw & Manolakis, 2002).

1.3.6. Hyperspectral Image and Hyperspectral Data

Hyperspectral image data consist of several congruent images representing intensities at different wavelength bands composed of vector pixels (*voxels*) containing two-dimensional spatial information (of m rows and n columns) as well as spectral information (of K wavelengths). These data are known as a three-dimensional hyperspectral cube, or *hypercube*, *datacube*, *data volume*, *spectral cube* or *spectral volume*, which can provide physical and/or chemical information of a material under test (Cogdill *et al.*, 2004). This information can include physical and geometric observations of size, orientation, shape, color, and texture, as well as chemical/molecular information such as water, fat, proteins, and other hydrogen-bonded constituents (Lawrence *et al.*, 2003). However, the combination of these two features (spectral and spatial) is not trivial, mainly because it requires creating a three-dimensional (3D) data set that contains many images of the same object, where each one of them is measured at a different wavelength. Because pixels are digitalized gray values or intensities at a certain wavelength, they may be expressed as integers. Intensity values of a spatial image in the *hypercube* at one wavelength may have 8-bit gray values meaning that 0 is the black and 255 is the white. In more precise systems, the intensity values of each pixel having 12-bit (2^{12} gradations, i.e., 0–4095), 14-bit (2^{14} gradations, i.e., 0–16383) or 16-bit (2^{16} gradations, i.e., 0–65535) gray levels are used. For many applications, 12-bit dynamic range is adequate and can provide

high frame rates. For more demanding scientific applications such as cell, fluorescence or Raman imaging, a higher performance 16-bit cooled camera may be advantageous.

Figure 1.2 illustrates one example of the *hypercube* extracted from a hyperspectral image acquired for a piece of meat. The raw hyperspectral image consists of a series of contiguous sub-images; each one represents the intensity and spatial distribution of the tested object at a certain waveband. All individual spatial images could be picked up from the *hypercube* at any wavelength(s) covering the spectral sensitivity of the system. Therefore, a *hyperspectral image* described as I*(x, y, λ)* can be viewed either as a separate spatial image I*(x, y)* at each wavelength *(λ)*, or as a spectrum I*(λ)* at every pixel *(x, y)*. Each pixel in a hyperspectral image contains the spectrum of that specific position. The resulting spectrum acts like a fingerprint which can be used to characterize the composition of that particular pixel. Since hyperspectral imaging acquires spatially distributed spectral responses at pixel levels, this allows flexible selection of any regions of interest on a target object,

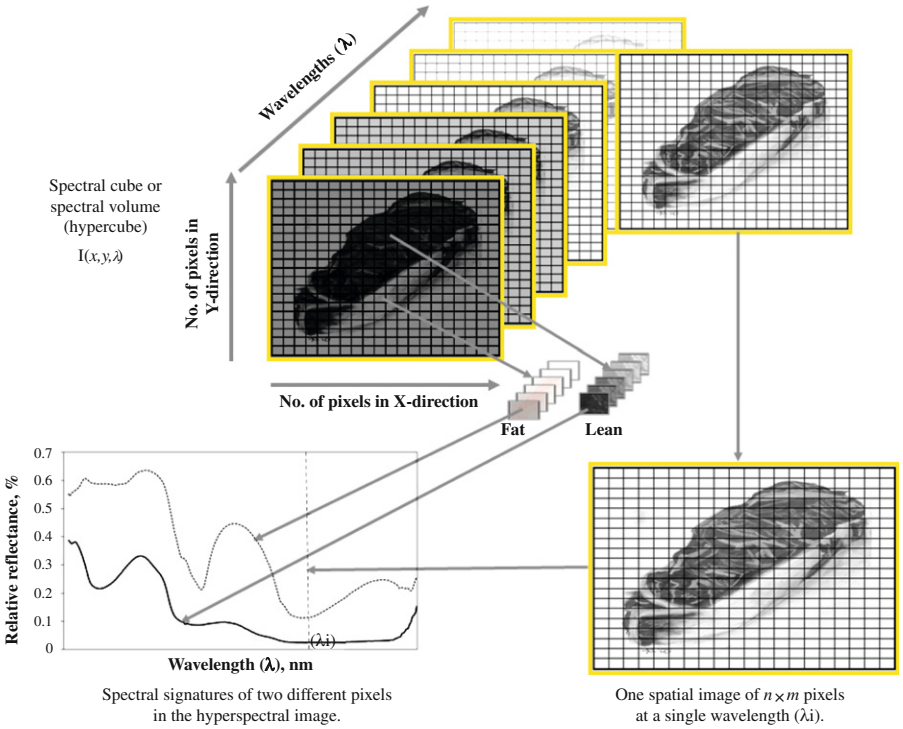

FIGURE 1.2

*Schematic diagram of hyperspectral image (*hypercube*) for a piece of meat showing the relationship between spectral and spatial dimensions. Every pixel in the hyperspectral image is represented by an individual spectrum containing information about chemical composition at this pixel. (Full color version available on http://www.elsevierdirect.com/companions/9780123747532/)*

e.g. variable sizes and locations. For instance, if two different pixels from two different compositional locations in the *hypercube* are extracted, they will show different fingerprints or different *spectral signatures*. Therefore, without any further manipulation or preprocessing treatments of these spectral data, the difference in spectral signatures between lean meat pixel and fat pixel of the tested piece of meat shown in Figure 1.2 are noticeably distinguished.

Technically speaking, the hyperspectral data are characterized by the following features:

Hyperspectral data volumes are very large and suffer from colinearity problems. This has implications for storage, management, and further image processing and analyses. The amount of data is the greatest problem that has to be coped with. Assuming collection of an image of 160 wavebands between 900 and 1700 nm (with 5 nm bandwidth) with spatial dimensions of 512×512 pixels and 8 bits precision (1 byte), the size of the image would be $512 \times 512 \times 160$ bytes $= 41.94$ Mega bytes. The primary goal of data analysis is therefore a reduction step to decrease the data size.

Hyperspectral data are inherently high dimensional since they are, by definition, composed of large numbers of spectral bands. For example, the hyperspectral imaging system that ElMasry *et al.* (2009) used in their experiment for chilling injury detection in apples and for predicting quality attributes in strawberries (ElMasry *et al.*, 2007) recorded 826 spectral bands in the VIS and NIR region between 400 and 1000 nm with about 0.73 nm between contiguous bands. Even though these high *dimensionality* data offer access to rich information content they also represent a dilemma in themselves for data processing especially when the major purpose is to use the system in a real-time application.

The *hypercube* can be viewed in the spatial domain as images ($m \times n$) at different wavelengths or in the spectral domain as spectral vectors at all wavelengths, as shown in Figure 1.3. Both representations are essential for analyzing the hyperspectral data with the suitable *chemometric* tools using one or more of the *multivariate analysis* techniques. For instance, if one hyperspectral image has dimensions of $256 \times 320 \times 128$, this *image cube* can be interpreted as 128 single channel images each with 256×320 pixels. Alternatively, the same hypercube can be viewed as 81,920 spectra, each with 128 wavelength points. This huge amount of data poses data mining challenges, but also creates new opportunities for discovering detailed hidden information.

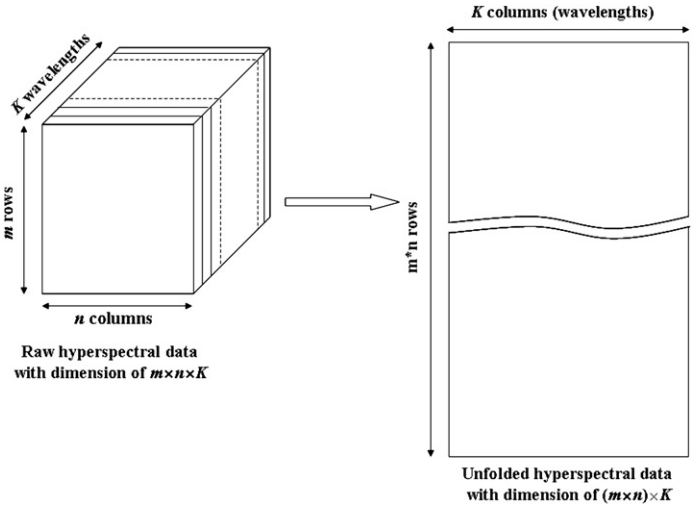

FIGURE 1.3 *Unfolding the hyperspectral data "hypercube" to facilitate multivariate analysis*

As explained in the previous sections, the product of a spectral imaging system is a stack of images of the same object, each at a different spectral narrow band. However, the field of spectral imaging is divided into three techniques called *multispectral*, *hyperspectral*, and *ultraspectral*. The concept of multispectral, hyperspectral, and ultraspectral imaging is similar. It is believed by many researchers that the only difference between them is the number of wavebands used during image acquisition. If an image is acquired with very few separated wavelengths, the system is called *multispectral imaging*. If the spectral image is acquired with an abundance of contiguous wavelengths, the system is then called *hyperspectral imaging*. While no formal definition exists, the difference is not based on the number of bands, contrary to various popular notations by many scientists working in this field. Multispectral deals with several images at discrete and somewhat narrow bands. The simplest method to obtain images at a discrete wavelength region is by using band-pass filters (or interference filter) in front of a monochrome camera lens. Multispectral images can be obtained by capturing a series of spectral images by using either a liquid crystal tunable filter (LCTF) or an acousto–optic tunable filter (AOTF), or by sequentially changing filters in front of the camera (Chen *et al.*, 2002). Regrettably, multispectral images do not produce the "spectrum" of an object. On the other hand, hyperspectral deals with imaging at narrow bands over a contiguous wavelength range, and produces the "*spectra*" of all pixels in the scene. Therefore a system with only 20 wavebands can also be

a hyperspectral system if it covers a certain spectral range (VIS, NIR, SWIR, IR, etc.) to produce spectra of all pixels within this range. Given that the visible range spectrum spans a wavelength range of approximately 300 nanometres (400–700 nm), the system of only 20 wavebands of 15 nm bandwidth can be named as hyperspectral. *Ultraspectral imaging* is typically used for spectral imaging systems with a very fine spectral resolution. These systems often have a low *spatial resolution* of several pixels only.

1.4. CONFIGURATION OF HYPERSPECTRAL IMAGING SYSTEM

The optical and spectral characteristics of a hyperspectral imaging system are determined largely by the application requirements. However, all systems have the same basic components in common: a means to image the object, a means to provide both spectral and spatial resolution, and a means to detect. The complete optical system for a hyperspectral imaging system consists of a suitable objective lens matched to the spatial and spectral requirements of the application, a wavelength dispersion device such as an imaging spectrograph and a two-dimensional detector such as a CCD or CMOS camera to simultaneously collect the spectral and spatial information. The main part of this system is the spectrograph. A spectrograph is a system for delivering multiple images of an illuminated entrance slit onto a photosensitive surface (detector). The location of the images is a function of wavelength. It is normally characterized by an absence of moving parts.

1.4.1. Acquisition Modes of Hyperspectral Images

There are three conventional ways to build one spectral image: area scanning, point scanning, and line scanning. These instruments capture a one- or two-dimensional subset of the datacube, and thus require the temporal scanning of the remaining dimension(s) to obtain the complete datacube. The *area-scanning* design, also known as *staring imaging* or *focal plane scanning imaging* or the *tunable filter*, involves keeping the image field of view fixed, and obtaining images one wavelength after another, therefore it is conceptually called the *wavelength-scanning* method or band sequential method. Acquiring an image at different wavelengths using this configuration requires a tunable filter, and the resulting hypercube data is stored in Band Sequential (BSQ) format. The *point-scanning* method, also known as *whiskbroom*, produces hyperspectral images by measuring the spectrum of a single point

and then the sample is moved and another spectrum is taken. Hypercube data obtained using this configuration are stored as Band Interleaved by Pixel (BIP) format. The third method is *line scanning*, also called *pushbroom*, involving acquisition of spectral measurements from a line of sample which are simultaneously recorded by an array detector; and the resultant hypercube is stored in the Band Interleaved by Line (BIL) format. This method is particularly well suited to conveyor belt systems, and may therefore be more practicable than the former ones for food industry applications.

In point scanning the sample is moved in the *x* and *y* directions point-by-point using a computer-controlled stage; meanwhile it is moved line-by-line in the case of line scanning. In imaging by area scanning, data are collected with a two-dimensional detector, hence capturing the full desired field-of-view at one time for each individual wavelength, without having to move the sample. The point-scanning and line-scanning methods are conceptually called *spatial-scanning* methods since they depend on scanning the specimen in the spatial domain by moving the specimen either point-by-point or line-by-line respectively, while area scanning is a spectral-scanning method. These three configurations of acquisition modes—based on the spectral imaging sensors—are explained in more detail below.

1.4.1.1. Staring imaging (area-scanning imaging, focal plane-scanning imaging or tunable filter or wavelength scanning)

The detector in an *area-scanning imaging* configuration is located in a plane parallel to the surface of the sample and the sample is imaged on the focal plane detector. The camera, lens, spectrograph, and the sample itself (field of view) remain fixed in position relative to the detector. The spectral domain is electronically scanned and the image is collected one spectral plane (wavelength) after another. One of the simplest methods for gathering the images at one wavelength at a time can be performed by collecting images using interchangeable narrow bandpass interference filters at distinct wavelengths. The bandpass size of the filters determines the number of wavelengths in the spectral range. The filters are positioned in front of the camera and a filter wheel rotates a bandpass filter into the optical path to acquire wavelength bands of equal bandwidth. This technique is usually preferred only where a limited number of wavebands are required because this process is inherently slow, which is considered one of its disadvantages. The disadvantage of using this configuration is the requirement for repetitive scanning of the same specimen at several wavelengths. Such repetition in scanning is necessary so that successive images at each wavelength increment can be

gathered. An alternative mechanism for obtaining wavelength scanning is to use *tunable filters*. Typically, this is achieved by using electronically tunable filters or imaging interferometers. In this configuration, the most predominantly employed filters are Liquid Crystal Tunable Filters (LCTFs), Acousto–Optic Tunable Filters (AOTFs), and interferometers either between the illumination source and specimen or between the specimen and the detector. The staring image acquisition is suitable for many applications where a moving tested sample is not required, such as florescence imaging using an excitation–emission matrix in which the wavelengths of both excitation and emission are controlled by the tunable filters where the filter change is done electronically. Lengthy image acquisition times can also be an issue for biological samples, which may be sensitive to heating caused by the continuous illumination from source lamps. Furthermore, staring imaging is not effective for either a moving target or for real-time delivery of information concerning a particular specimen.

1.4.1.2. Whiskbroom (point-scan imaging or Raster-scanning imaging)

It is obvious that the easiest way to acquire a particular spectral image of an object is to use a filter-based imaging system (i.e., area-scanning imaging). This is mostly due to the poor optical quality and transmission efficiency of wavelength dispersive systems such as those based on a diffraction *grating*. The use of newer, highly specialized *prism* spectrometers has enabled the design of spectral imaging systems with high efficiency. The *whiskbroom* is an example of this technology which operates as an electromechanical scanner with a single detector. Whiskbroom scans a single pixel at a time, with the scanning element moving continuously. Light coming from the specimen is dispersed using an optical grating, prism or a similar *dispersing element* and is detected wavelength by wavelength by a line detector array. Thus whiskbroom scanners have one detector element for each wavelength (spectral band) recorded. A single, small sensor can be moved in a zigzag or raster fashion to sense the light intensity on a grid of points covering the whole image. The image is recorded with a double scanning step: one in the wavelength domain and the other in the spatial domain. This design is commonly used for microscopic imaging where the acquisition time is usually not a problem since a double scan (i.e., spatial and spectral) is required. By moving the sample systematically in two spatial dimensions, a complete hyperspectral image can be obtained. This system provides very stable high resolution spectra; however, positioning the sample is very time-consuming and has high demands on repositioning hardware to ensure

repeatability. The spatial size dimensions of the hyperspectral image are limited only by the sample positioning hardware.

1.4.1.3. Pushbroom (line-scan imaging)

Line-scanning devices record a whole line of an image rather than a single pixel at a time using a two-dimensional *dispersing element* (*grating*) and a two-dimensional *detector* array. A narrow line of the specimen is imaged onto a row of pixels on the sensor chip and the spectrograph generates a spectrum for each point on the line, spread across the second dimension of the chip. Therefore, hyperspectral images are acquired by a wavelength dispersive system that incorporates a diffraction *grating* or *prism*. These instruments typically require an entrance *aperture*, usually a *slit*, which is imaged onto the focal plane of a spectrograph at each wavelength simultaneously. Therefore, an object imaged on the slit will be recorded as a function of its entire spectrum and its location in the sample. In this design an array of detectors is used to scan over a two dimensional scene using a two dimensional detector perpendicular to the surface of the specimen. This configuration is normally used when either the specimen or the imaging unit is moving one in respect to the other, such as those used in industrial applications. The sensor detectors in a pushbroom scanner are lined up in a row called a linear array. Instead of sweeping from side to side as the sensor system moves forward, the one-dimensional sensor array captures the entire scan line at once. Since no filter change is required, the speed of image acquisition is limited only by camera read-out speeds.

The difference between *wavelength scanning* (implemented in tunable filter systems) and *spatial scanning* (implemented in pushbroom systems) approaches to acquire a cube of spatial and spectral data is shown in Figure 1.4. One approach is used to acquire a sequence of two-dimensional images at different wavelengths (from λ_1 to λ_n) and the other approach is used to acquire a sequence of line images in which a complete spectrum is captured for each pixel on the line. In the first approach (*wavelength scanning*), illustrated in Figure 1.4a, the detector sequentially captures a full spatial scene at each spectral band (wavelength) to form a three-dimensional image cube. This approach is preferable if the number of bands needed is limited and the object can be held fixed in front of the camera during capturing. In the second approach (*spatial scanning*), shown in Figure 1.4b, a line of spatial information with a full spectral range per spatial pixel is captured sequentially to complete a volume of spatial–spectral data (Kim *et al.*, 2001). Since the *spatial-scanning* mode requires moving the specimen line by line, this method is particularly well suited to conveyor belt systems

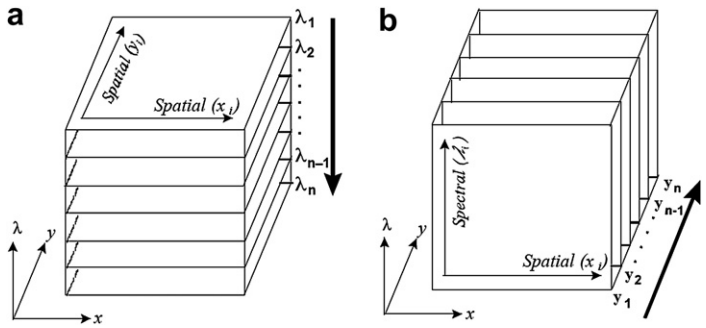

FIGURE 1.4 *Conceptual representations of image acquisition modes. Data arrows indicate directions for sequential acquisition to complete the volume of spatial and spectral data "hypercube". (a) Wavelength-scanning mode; (b) spatial-scanning mode*

and is more practicable than the *wavelength scanning* for real-time applications (Chen *et al.*, 2002; Mehl *et al.*, 2004; Polder *et al.*, 2002).

1.4.2. Detectors in Hyperspectral Imaging Systems

The two-dimensional *detector* (i.e., the area detector) for the spectrograph of the hyperspectral imaging system plays an important role in recording the spatial and spectral signals. The detectors used in hyperspectral imaging systems are generally photovoltaic semiconductor detectors, so-called *charge-coupled devices* (CCDs). Semiconductor devices are electronic components that exploit the electronic properties of semiconductor materials, principally silicon (Si), germanium (Ge), and gallium arsenide (GaAr). Silicon (Si) is the most widely used material in semiconductor devices. The many advantages such as low raw material cost, relatively simple processing, and a useful temperature range makes it currently the best compromise among the various competing materials. Semiconductor line or area arrays typically used in most spectral imaging systems include silicon (Si) arrays, indium antimonide (InSb) arrays, mercury cadmium telluride (HgCdTe) arrays, and indium gallium arsenide (InGaAs) arrays. Silicon arrays are sensitive to radiation in the 400–1000 nm wavelength range, InSb, HgCdTe, and InGaAs arrays at longer wavelengths between 1000 and 5000 nm. In some instruments, several different and overlapping detector elements are used for optimized sensitivity in different wavelength regions (Goetz, 2000). To increase detection efficiency especially in the infrared regions, the detector should be cooled. Cooling reduces the array's dark current, thus improving the sensitivity of the detector to low light intensities, even for ultraviolet and visible wavelengths, and hence reducing the thermal noise to a negligible level.

1.4.3. Main Components of Hyperspectral Imaging System

In food analysis applications it is desirable to know what is the main components of the most acceptable hyperspectral system in this field. Therefore, in this section the main components of a hyperspectral imaging system employing the pushbroom design will be explained due to the fact that it uses the line-scan method and therefore is more consistent for on-line application. An image of a specimen located in the field of view (FOV) is collected by translating the specimen across the slit aperture of the spectrograph in a pushbroom acquisition method. Thus the spectral data are measured simultaneously and the image or FOV is generated sequentially. The prime advantage of this method is that all the wavelength data needed to identify an object or objects, even if the spectra are highly convoluted, are acquired simultaneously and are immediately available for processing. Consequently, this technique is ideal for kinetic studies on samples that exhibit movement, for studies of time-based changes in molecular characteristics, and for any condition that benefits from real-time spectral analysis. As stated by many researchers (e.g. Kim *et al.*, 2002; Polder *et al.*, 2002), the pushbroom hyperspectral imaging system consists of five main components: camera containing a cooled two-dimensional (2D) light detector, spectrograph, translation stage, illumination units, and a computer. Each of these components has its own characteristics that influence the total accuracy of the system. To characterize the performance of the whole system, it is important to measure and optimize all parameters that influence the quality of the obtained spectral image. For instance, the ideal illumination should be homogeneous illumination over a large area without radiation damage to the samples. By scanning the object by moving the linear translation stage, the second spatial dimension is incorporated, resulting in a three-dimensional (3D) datacube of (x, y, K) dimensions. The main components of a pushbroom hyperspectral imaging system used for nondestructive meat quality assessment in University College Dublin (UCD), Ireland, are depicted in Figure 1.5.

The wavelength dispersing unit in the hyperspectral imaging system is essentially a grating spectrograph with a 2D detector array. It utilizes a field-limiting entrance slit and an imaging spectrometer with a dispersive element to allow the 2D detector to sample the spectral dimension and one spatial dimension simultaneously. The imaging lens focuses the light onto an entrance slit, the light is then collimated, dispersed by a grating and focused on the detector. The second spatial dimension, y, is typically generated by moving or scanning the camera's field of view relative to the scene. The spectral resolution of the system depends on both the slit width and the

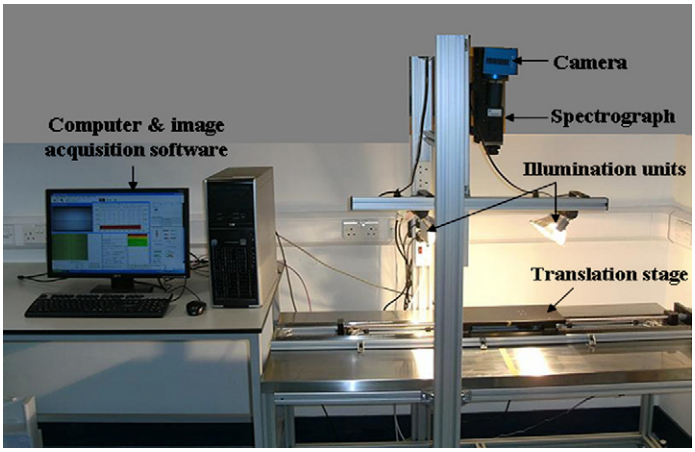

FIGURE 1.5 *Main components of a pushbroom hyperspectral imaging system. (Full color version available on http://www.elsevierdirect.com/companions/9780123747532/)*

optical aberration. As the light beam enters the spectrograph, it is dispersed into different directions according to wavelength while preserving its spatial information. The dispersed light is then mapped onto the detector array, resulting in a 2D image, one dimension representing the spectral axis and the other containing the spatial information for the scanning line. By scanning the entire surface of the specimen, a complete 3D hyperspectral image cube is created, where two dimensions represent the spatial information and the third represents the spectral information (Lu, 2003b). Figure 1.6 shows an implementation of this principle from Specim Ltd (Finland).

Technically speaking in the context of system integration, the basic elements of a hyperspectral imaging spectrograph are shown in Figure 1.7. The light source, such as a halogen lamp, illuminates the object to be measured, and the entrance optics, e.g. a camera lens, collects the radiation from the object and forms an image on the image plane (image plane 1 in Figure 1.7), where the entrance slit of the imaging spectrograph is located. The slit acts as a field-stop to determine the instantaneous FOV in spatial directions to a length of $\triangle x$ and a width of $\triangle y$, marked as the measured area in Figure 1.7. Each point A in the spatial **x**-direction of the measured area has its image A′ on the entrance slit. The radiation from the slit is collimated by either a lens or a mirror and then dispersed by a dispersing element, which is typically a prism or grating, so that the direction of propagation of the radiation depends on its wavelength. It is then focused on image plane 2 by the focusing optics, i.e. a lens or mirror. Every point A is represented on image plane 2 by a series of monochromatic images forming a continuous spectrum

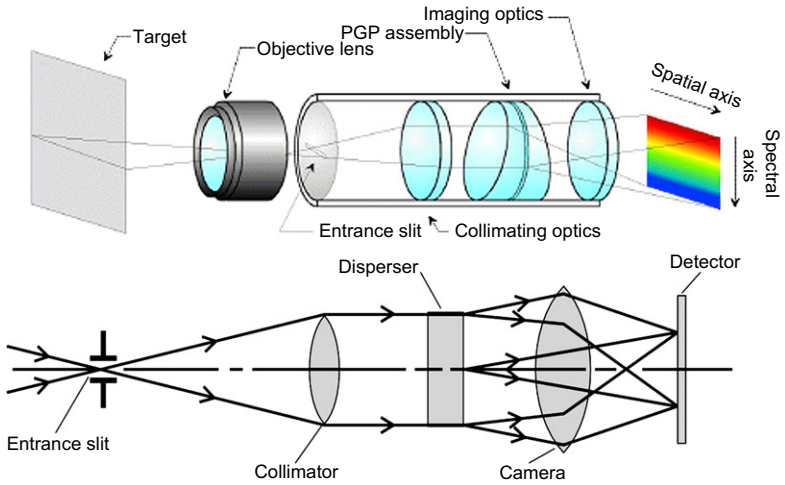

FIGURE 1.6 *Working principle of prism-grating-prism (PGP) spectrograph (courtesy of Specim Ltd). (Full color version available on http://www.elsevierdirect.com/companions/9780123747532/)*

in the direction of the spectral axis, marked with different sizes of A″. The focused radiation is detected by a 2D detector array such as charge-coupled device (CCD) or a complementary metal-oxide-semiconductor (CMOS) detector. The imaging spectrograph allows a 2D detector array to sample one spatial dimension of length $\triangle x$ and infinite width $\triangle y$ and the spectral dimension of the 3D cube simultaneously. The width $\triangle y$ also defines the spectral resolution, which can be seen as $\triangle y''$ in the direction of the spectral

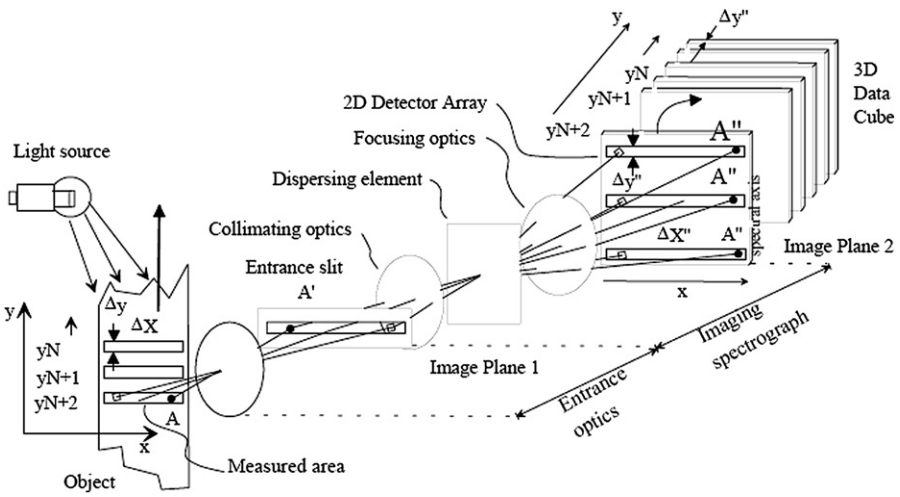

FIGURE 1.7
The basic elements of a hyperspectral imaging spectrograph, with the entrance optics and generation of the 3D datacube: spatial (x and y) and spectral (K) dimensions (reproduced from Aikio, 2001 by permission of the author)

axis in Figure 1.7. In addition to defining spectral resolution, slit width controls the amount of light entering the spectrograph. Also, the collimator makes this light parallel so that the disperser (a grating or prism) disperses it. The second spatial dimension of the object, y, is generated by scanning or moving the FOV of the instrument relative to the scene, corresponding to the positions $y\mathrm{N}$, $y\mathrm{N}+1$, $y\mathrm{N}+2$ in Figure 1.7.

1.5. CALIBRATION OF HYPERSPECTRAL IMAGING SYSTEM

Hyperspectral imaging systems are basically able to delineate multiple mapping of essential chemical constituents such as moisture, fat, and protein on most biological specimens by performing spectral characterizations of these constituents. However, some systems may present inconsistent spectral profiles of reference spectra even under controlled conditions. This variability confirms that there is a need for a standardized, objective *calibration* and a validation protocol. In all hyperspectral imaging systems the *spectrograph* and its dispersive element is the most important component for the determination of its optical properties because it determines the spectral range and the spectral resolution. The dispersive element separates the light depending on its wavelengths and projects these fractions on different spatial positions. Therefore, the goals of the calibration process are to (a) standardize the spectral axis of the hyperspectral image, (b) determine whether a hyperspectral imaging system is operating properly, (c) provide information about the accuracy of the extracted spectral data and thus validate their acceptability and credibility, and (d) diagnose instrumental errors, measurement accuracy, and reproducibility under different operating conditions.

In essence, calibrating a spectral imaging system is vital before acquiring the images. A system calibration test is always a prudent step when doing qualitative and quantitative analyses. This procedure is performed after assembling all the components of the hyperspectral imaging system to ensure both spectral and spatial dimensions are projected in their right directions. The manufacturers are obliged to produce calibrated systems to guarantee trustworthy results. Recalibration is generally not required unless the physical arrangement of the components of the imaging system is disturbed. The first precaution in the calibration process is to cool the imaging system to its initial operating temperature, which is usually between -80 and $-120\,^{\circ}\mathrm{C}$ in most modern systems. Also, the combination of lamp

intensity and detector integration time has to be adjusted to avoid saturation of the analog to digital (A/D) converter.

Another precaution that requires consideration is to set image binning, which is determined by the spectral distribution of useful wavelengths and the size of spatial image features to be processed for the application. In the case of line-scanning mode (pushbroom), one of the dimensions is assigned to one spatial axis and the other is used for projecting the spectral axis as a spectral dispersion plane. For instance, if the image resolution is of $x \times y$ pixels, x pixels will be used for projecting the spatial resolution of the scanned line and y pixels will be used for projecting the spectral resolution of K wavelengths. Moreover, wavelength dispersion controls the physical distance that separates one wavelength from another on the spectral axis and is a key parameter in determining the limits of spectral resolution. The binning in both spatial and spectral directions will lead to a reduction in the resolution of both axes. The new resolution will be the initial number of pixels of this axis over the binning factor. Therefore, the new resolution would be x/b_1 pixels for spatial resolution and y/b_2 pixels for the spectral resolution, where b_1 and b_2 are the binning factors in the spatial and spectral axes respectively. To make this sophistication much clearer, it can be considered that the spatial and spectral resolution in most widely used hyperspectral imaging systems implemented in food quality assessment are of 512×320 pixels. If under certain applications a unity binning factor ($b_1 = 1$) is required in the spatial direction, this will result in line-scan images with a spatial resolution of 512 pixels (512 divided by 1). On the other hand, if a binning factor of value $b_2 = 2$ is used the resulting spectral resolution would be 160 pixels (320 divided by 2) in the spectral axis. This will lead to a total number of 160 contiguous wavebands (channels) in the spectral axis. Strictly speaking, the binning process in the spectral direction adds together photons from adjacent pixels in the detector array which will produce a reduced number of pixels to be digitized by the A/D converter for the computer to process. Reducing total pixel readout time decreases the acquisition time of each line-scan image, which allows a higher image acquisition speed for the imaging device.

The most significant step in the calibration process is the spectral waveband calibration (wavelength calibration) that identifies each spectral channel with a specific wavelength. Each wavelength on the spectral axis is identified as a function of its physical location on this axis. To determine the relation between distance (in pixels) on the spectral axis and wavelength, the spectral axis must be calibrated by using a standard emission lamp as a light source. A specific wavelength will then be assigned to a specific column of CCD pixels. The most acceptable calibration protocol involves the use of a single or multi-ion discharge lamp of mercury (Hg), helium (He), argon (Ar),

neon (Ne), and/or cadmium (Cd) that emits distinct, stable, spectral features in place of a sample. These reference spectra from this lamp will be used to accurately predict the spectral resolution of the system and adjust the spectral axis. Therefore, using these reference light sources that emit absolute standard ''reference spectra'' is a sensible tool for diagnosing instrumental errors and measurement accuracy and reproducibility under different operating conditions. With this information on one hand, the researcher can determine whether the spectral imaging system is working optimally and make objective comparisons with the performance of other spectral imaging systems. On the other hand, if spectral imaging systems are standardized to produce the same spectral profile of a reference lamp, the researcher can be confident that the experimental findings are comparable with those obtained from other spectral imaging systems. Different light sources of known spectrum should be used for this task, such as mercury, helium, and/or cadmium calibration lamps, as shown in Figure 1.8. One example of a single ion discharge calibration lamp is the cadmium lamp that has five distinct peaks in the visible range of the electromagnetic spectrum at 467.8, 479.9, 508.58, 607.2, and 643.8 nm. as depicted in Figure 1.8.

In addition, there are several readily available calibration sources of a multi-ion discharge type, the most common of which is a low-pressure Hg^{+}/Ar^{+} discharge lamp that covers the wavelength range of 400 to 840 nm. The emission spectrum of this lamp is shown in Figure 1.9 (Oriel Instruments, Stratford, CT, USA). The benefit of this spectrum is that the spectrum acts as a spectral fingerprint that can be used to calibrate the performance of any spectroscopic system.

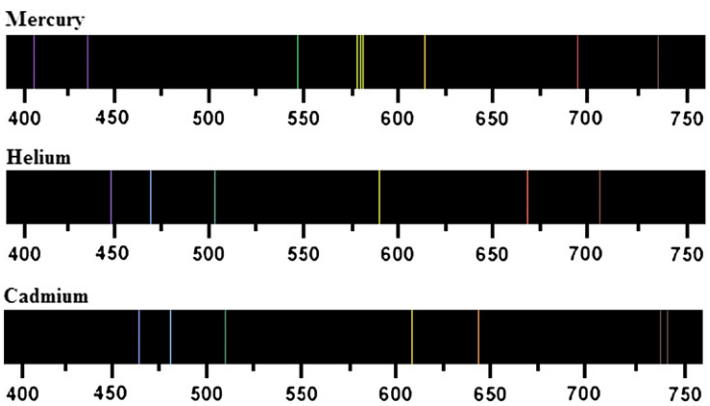

FIGURE 1.8 *Emission (bright lines) spectra of different calibration lamps. (Full color version available on http://www.elsevierdirect.com/companions/9780123747532/)*

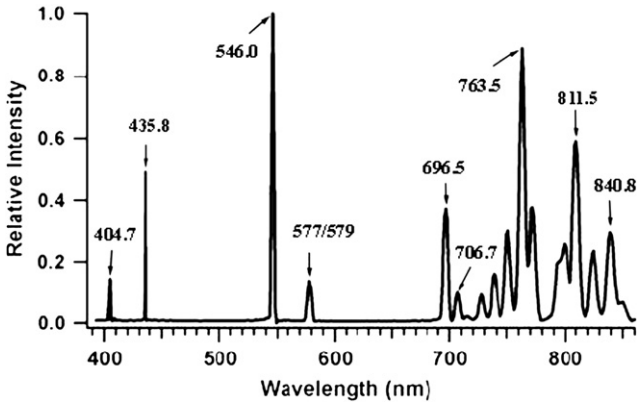

FIGURE 1.9 *Spectrum of calibration light source of pure Hg^+/Ar^+ low-pressure discharge lamp*

In practice, the calibration lamp is first scanned by the hyperspectral imaging system under controlled operating conditions. Once the calibration lamp is scanned, its peaks are then assigned to standardize the spectral axis. Then, a polynomial regression of first or second order can be established to convert spectral axis (in pixels) to its corresponding wavelength using the reference wavelength peaks of the calibration lamp. Following system calibration, the spectral imaging system will be ready to use for the acquisition of real line-scan images. The extracted data from such images before calibration will be in the pure form (nonlinearized pixel versus intensity), and after calibration will be wavelength versus intensity. However, if some error occurs in the physical arrangement of the hyperspectral imaging system or if some of its components have to be reassembled, the system should be recalibrated with the calibration lamp. The system can be used safely provided that it gives the same peaks of the calibration lamp with an acceptable error. This step must be repeated several times to diagnose the level of this error and to judge the reproducibility of the system under different operating conditions.

Finally, after acquiring hyperspectral images of real samples, another calibration step, called *reflectance calibration*, should be performed to account for the background spectral response of the instrument and the 'dark' current of the camera. The background is obtained by acquiring a spectral image from a uniform, high reflectance standard or white ceramic (~100% reflectance), and the dark response (~0 % reflectance) is acquired by recording an image when the light source is turned off and the camera lens is completely covered with its nonreflective opaque black cap. These two

reference images are then used to calculate the pixel-based relative reflectance for the raw line-scan images using the following formula:

$$I = \frac{I_0 - D}{W - D} \tag{1.3}$$

where I is the relative reflectance image, I_0 is the raw reflectance image, D is the dark reference image, and W is the white reference image.

The corrected hyperspectral image can also be expressed in absorbance (A) by taking logarithms of the above equation as:

$$A = -\log_{10}\left(\frac{I_0 - D}{W - D}\right) \tag{1.4}$$

1.6. SPECTRAL DATA ANALYSIS AND CHEMOMETRICS

Hyperspectral imaging systems cannot stand alone without the help of some software for gaining high performance in acquisition, controlling, and analyses. It is essential to support the system with software for image acquisition, software for controlling the motor to move the sample line by line, software for extracting spectral data and preprocessing steps, software for multivariate analysis, and software for final image processing. Integration of image acquisition, spectral analysis, chemometric analysis, and digital image analysis in single software has not been explored yet. In fact, some of these processes are integrated in one software package to perform some of these operations. Alternatively, professional researchers can develop their own software routines or build a comprehensive graphical user interface (GUI) to perform each of the key steps of these processes. Typically, routines can be developed by using packages that support scripting capability, such as C^{++}, Matlab, IDL or LabView. However, researchers should be familiar with the main fundamentals of the necessary steps required to obtain the key information about the process or about the samples being monitored for achieving the final goals of the tests. Typical steps usually undertaken in hyperspectral imaging experiments are outlined in the flowchart described in Figure 1.10.

The first step is the collection of a hyperspectral image by utilizing ideal acquisition conditions in terms of illumination, spatial and spectral resolution, motor speed, frame rate, and exposure/integration time. After acquiring a hyperspectral image for the tested sample, this image should be calibrated with the help of white and dark hyperspectral images as mentioned earlier in

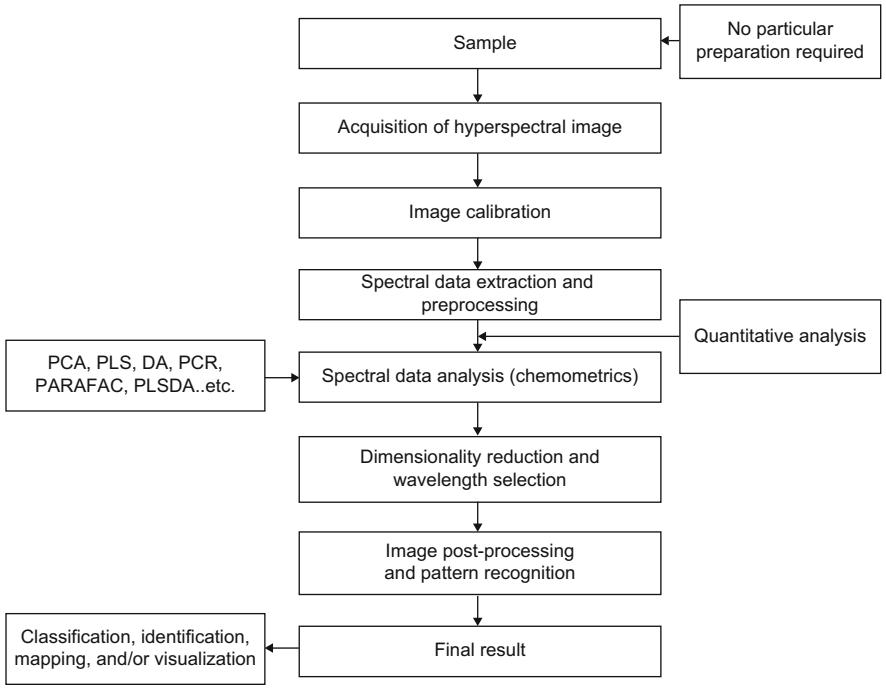

FIGURE 1.10 *Flowchart of the key steps involved in hyperspectral imaging analyses*

this chapter. The spectral data are then extracted from different regions of interest (ROIs) that present different quality features in the calibrated image. Extracted spectral data should be preprocessed to reduce noise, improve the resolution of overlapping data, and to minimize contributions from imaging instrument responses that are not related to variations in the composition of the imaged sample itself. *Preprocessing* of spectral data is often of vital importance if reasonable results are to be obtained from the spectral analysis step. Preprocessing includes spectral and spatial operations. Spectral preprocessing includes some operations such as spectral filters, normalization, mean centering, auto scaling, baseline correction, differentiation (Savitsky-Golay), standard normal variate (SNV), multiplicative scatter correction (MSC), and smoothing. On the other hand, spatial operations include low-pass filters, high-pass filters, and a number of other spatial filters. Detailed overviews of the most admired preprocessing operations are further explained in subsequent relevant chapters in the book.

Once instrument response has been suppressed by means of preprocessing, qualitative analysis can be employed. Qualitative analysis attempts to address what different components are present in the sample and how these

components are distributed. Many *chemometric* tools fall under this category. Strictly speaking, the cornerstone of this process is the data analysis using multivariate analysis by one or more chemometrics tools, including correlation techniques such as cosine correlation and Euclidean distance correlation; classification techniques such as principal components analysis (PCA), cluster analysis, discriminant analysis (DA), and multi-way analysis; and spectral deconvolution techniques. To build concentration maps for determining the estimated concentrations of different components present in the tested sample and their spatial distribution, a quantitative assessment should be performed using a standard analytical means. In quantitative spectral analysis, a number of multivariate chemometric techniques can be used to build the calibration models to relate spectral data to the actual quantitative data. Depending on the quality of the models developed, the results can range from semi-quantitative concentration maps to rigorous quantitative measurements.

Moreover, with the aid of multivariate analysis, the huge *dimensionality* and *colinearity* problems of hyperspectral data can be reduced or eliminated by selecting the spectral data at some important wavelengths. In most cases, not all the spectral bands are required to address a particular attribute. Selection of *important wavelength* is an optional step based on the speed requirements of the whole process. Generally, the selection of these optimal wavelengths reduces the size of the required measurement data while preserving the most important information contained in the data space. The wavelength preserving the largest amount of energy among the hyperspectral data carries the most important spectral information and maintains any valuable details about the tested samples. The selected *essential wavelengths* should not only maintain any valuable required details, but also simplify the successive discrimination and classification procedures (Cheng *et al.*, 2004). Indeed, the selection of the most *efficient wavelength* can be done off-line and then the on-line process consisting of image acquisition and analyses may be executed at acceptable speeds (Kleynen *et al.*, 2005). Several essential wavelengths could be sorted from the whole spectral cube through a variety of strategies, such as general visual inspection of the spectral curves and correlation coefficients (Keskin *et al.*, 2004; Lee *et al.*, 2005), analysis of spectral differences from the average spectrum (Liu *et al.*, 2003), stepwise regression (Chong & Jun, 2005), discriminant analysis (Chao *et al.*, 2001), principal component analysis (PCA) (Mehl *et al.*, 2004; Xing & De Baerdemaeker, 2005), partial least square (PLS), and others (ElMasry *et al.*, 2009; Hruschka, 2001). The mathematical principles of these approaches are given in subsequent relevant chapters in the book.

Results obtained from preprocessing, qualitative analysis, and quantitative analysis must be visualized either by scaling, surface mapping or pseudocolor representation. Once the final digital concentration images have been generated, traditional postprocessing of these images, such as segmentation, enhancement, and morphological feature extraction can be applied as a final step of the work flow. The final image processing step is carried out to convert the contrast developed by the classification step into a picture depicting component distribution. Grayscale or color mapping with intensity scaling is commonly used to display compositional contrast between pixels in an image. Final results of these calculations are used to develop key quantitative image parameters to characterize various traits in the tested samples in different categories by performing classification, identification, mapping and/or visualization.

1.7. CONCLUSIONS

Hyperspectral imaging is a complex, highly multidisciplinary field that can be defined as the simultaneous acquisition of spatial images in many spectrally contiguous bands. It is quite clear that measurement in contiguous spectral bands throughout the visible, near-infrared and/or shortwave regions of the electromagnetic spectrum makes it possible to collect all the necessary information about the tested objects. Each pixel in the hyperspectral image contains a complete spectrum. Therefore hyperspectral imaging is a very powerful technique for characterizing and analyzing biological and food samples. The strong driving force behind the development of hyperspectral imaging systems in food quality evaluation is the integration of spectroscopic and imaging techniques for discovering hidden information nondestructively for direct identification of different components and their spatial distribution in food samples. As a result, hyperspectral imaging represents a major technological advance in the capturing of morphological and chemical information from food and food products. Although effective use of hyperspectral imaging systems requires an understanding of the nature and limitations of the data and of various strategies for processing and interpretation, the wealth of additional information available and the application benefits that hyperspectral imaging produce are almost without limit in monitoring, control, inspection, quantification, classification, and identification purposes. It is therefore anticipated that work in this area will gain prominence over the coming years and its potentialities present significant challenges to food technologists and food engineers.

NOMENCLATURE

Symbols

E	energy of the photon (J)
h	Planck's constant (6.626×10^{-34} J.s)
f	frequency (Hz)
c	speed of light in vacuum (299 792 458 ms^{-1})
υ	speed of the wave, ms^{-1} (equals c in a vacuum)
I	relative reflectance image (calibrated image)
I_0	raw reflectance image
D	dark reference image
W	white reference image
A	absorbance calibrated spectral image

Abbreviations

AM	amplitude modulation of radio waves
AOTF	acousto–optic tunable filter
BIL	band interleaved by line
BIP	band interleaved by pixel
BSQ	band sequential
CCD	charge-coupled device
CMOS	complementary metal-oxide-semiconductor
DA	discriminant analysis
FIR	far-infrared
FLIM	fluorescence lifetime imaging microscopy
FM	frequency modulation of radio waves
FOV	field of view
FWHM	full width at half maximum
HACCP	hazard analysis critical control point
IR	infrared
LCTF	liquid crystal tunable filter
MSC	multiplicative scatter correction
NIR	near-infrared
NIRS	near-infrared spectroscopy
PCA	principal component analysis
PCR	principal component regression
PLS	partial least square
RGB	red, green, blue (components of a color image)
ROI	region of interest
SNR	signal-to-noise ratio

SNV	standard normal variate
SWIR	shortwave-infrared
UV	ultraviolet
VIS	visible light

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