Current Horticulture 12(1): 13–22, Januray–April 2024 DOI: 10.48165/chr.2023.12.1.02

Hyperspectral imaging/reflectance as a tool for assessment of nutritional and quality-related parameters in tomato (*Solanum lycopersicum*) fruits - a review

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Received: 3 July 2021; Accepted: 8 March 2023

ABSTRACT

Tomato (*Solanum lycopersicum* L.) is most important vegetable crop for human health. The postharvest handling and management of tomato is prime concern in India because the annual postharvest losses for tomato can reach up to 25 - 40 %. Non-destructive approaches for quantification and monitoring of nutritional and quality aspects of horticultural commodities have come up in a big way in the recent past that can also serves towards better postharvest management. Out of various non-destructive approaches, optical method based on visible-near infrared (Vis-NIR) spectroscopy (hyperspectral imaging and reflectance) is the most important analytical tool that provides spatial and spectral information simultaneously for a commodity towards non-destructive assessment of food quality-related parameters. Therefore, an overview with latest developments and applications of hyperspectral imaging and reflectance techniques for assessment of nutritional and quality parameters of tomato fruits have been discussed. The advantages and disadvantages of this tool along with the future perspectives are also highlighted.

Key Words: Carotenoids, Firmness, Lycopene, Maturity, Non-destructive methods, Nutritional quality, Quality assessment, Reflectance based indices, Ripeness

Tomato (*Solanum lycopersicum* L.) fruit is most important vegetable in the world with wider consumption both in raw or in processed forms (Sharma *et al*., 2022). Tomato fruit is available throughout the year and it has known beneficial effects on human health (Ali *et al*., 2021; Collins *et al*., 2022). These compounds also serve as nutraceuticals due to their anti-oxidative, anti-carcinogenic and anti-mutagenic actions. Versatile health benefits emphasize the need of tomato fruits in our daily diet (Ramesh *et, al*., 2021a; b; Collins *et al*., 2022; Sharma *et. al*., 2022). However, tomato fruit is highly perishable due to its climacteric nature of fruit ripening (Paul *et, al*., 2012; Paul *et al*., 2014). Therefore postharvest losses reach up to 25 - 40 % (Paul and Pandey, 2016; Paul and Pandey, 2018). So, to reduce

The use of non-destructive approaches are advantageous because with the destructive measurements it is not possible to monitor changes with the progress of developmental, ripening and passage of time for the same fruit or fruit lot. In this context, non-destructive measurements are of immense importance. In addition to this, identification of exact ripening stage and quality status in a non-destructive way can also provide more reliable and meaningful information on fruit biology, fruit physiology and fruit ripening that in turn become key in deciding the time of harvesting, need for grading, transportability, potential shelf-life, overall storability and final quality aspects (Dale *et, al*., 2013; Tiwari *et al*., 2013; Paul *et, al*., 2018; Wang *et al*., 2021). Furthermore, dynamics of bioactive compounds present in fruit is also strongly related to progression of ripening and changes in physiological status. Changes in profile of bioactive compounds over a period of time can also form an applied perspective to a wide range of scientific investigations with practical applicability for physiological processes such as fruit ripening and ripening-related changes (Paul *et, al*., 2011; Sharma *et, al*., 2020).

Non-destructive methods with above merits will be highly desirable for monitoring, remote, online, mechanization and automation of handling, sorting, quality assessment and packaging of fresh commodities in economic, safe and environment-friendly way (Hussain *et al*., 2018; Wang *et al*., 2021). Various aspects related with supply chain of tomato including quality control, inspection, random sampling, selection, decision making and exportability can also get facilitated with the availability of such techniques/methods (Jiang *et al*., 2013; Wu and Sun 2013; Paul *et. al*., 2018). Out

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of various non-destructive methods, optical method based on visible-near infrared (Vis-NIR) spectroscopy (hyperspectral imaging and reflectance) is emerging as the most important analytical tool for assessment of food quality. This technique has been employed for estimation of various quality and other physiological parameters in a number of fruits and vegetables (Jha and Garg, 2010; Xiang *et al*., 2022; Hasanzadeh *et al*., 2022). This review presents an update on developments and applications of hyperspectral imaging/reflectance technique for the assessment of nutritional and quality parameters of tomato fruits. It also presents advantages and disadvantages of this technique along with the future perspectives.

Hyperspectral imaging and reflectance

All food samples continuously emit and absorb energy by lowering or raising their molecular energy levels when exposed to light or in presence of light. The wavelengths at which molecules absorb, reflect, and transmit electromagnetic radiations revel the characteristics of their structure and composition (Sun, 2010). Any biological material and food tissues are held together by several different molecular bonds and forces. Water, carbohydrates and fats are rich in O-H or C-H bonds. Organic compounds and their derivatives are rich in C-H or N-H bonds. When a food sample is exposed to light, electromagnetic waves are transmitted through it, the energy of incident electromagnetic waves [ranging from ultraviolet radiation (UV), visible light (Vis), near-infrared (NIR), mid-infrared (MIR) and far-infrared (FIR)] change because of the stretching and bending vibrations of chemical bonds such as O-H, N-H and C-H (Wang *et al*., 2021). The changes in molecular energy levels are able to tell *via* spectroscopy about the characteristic and detailed fingerprints of food samples. At the macro level, the electromagnetic waves are recorded as light and the transitioning of the incident electromagnetic waves emerge out as reflection, scattering, and transmission of light. Since the absorbed part of light penetrates into the tissue of samples, the strength and wavelengths of emission and absorption depend on the physical and chemical states of the objective material. The emerging light can be converted into a spectrum and reshaped into images by hyperspectrometers with high signal to noise ratios. The obtained images (hyperspectral images) and reflectance (hyperspectral reflectance) indicate the chemical constituents and physical properties of the food samples.

Hyperspectral imaging or reflectance, also known as chemical or spectroscopic imaging or reflectance, are the most powerful and fastest-growing non-destructive tools for food quality analysis and control (ElMasry and Sun, 2010; Wu and Sun, 2013). They integrate imaging and spectroscopy technologies into one system that provide both spatial and spectral information simultaneously from an object (commodity). Thus, hyperspectral imaging and reflectance has capability to monitor rapidly and non-invasively both, physical and morpho-physiological characteristics besides the intrinsic chemical and molecular information of a food product for the purpose of quality assessment and safety analysis. Thus, hyperspectral imaging/reflectance is a very powerful technique for characterizing and analyzing biological and food samples. Keeping in view of various merits and large number of applications of this technique, there has been an increasing interest in hyperspectral data acquisition with a focus to develop techniques/methods for analyzing spectra of plant part to quantify and then predict the concentrations of pigments, contents and physiological status of plant part under investigation.

Acquisition of hyperspectral data

There are three common sensing modes for hyperspectral imaging, namely; reflectance, transmittance and interactance as illustrated in Fig. 1 Positions of light source and the optical detector (cameral, spectrograph, and lens) are different for each of the acquisition mode.

Reflectance mode: Detector capture the reflected light from the illuminated sample in a specific conformation to avoid specular reflection (Fig. 1a). External quality features can be detected using reflectance model which include size, shape, colour, surface texture and external defects etc. Reflectance mode can be used for thicker samples. It is therefore that food materials can be inspected as a whole in reflectance mode without the need to make slices.

***Transmittance mode*:** Here detector is located in the opposite side of the light source (Fig. 1b), it captures the transmitted light through the sample which carries more valuable internal information but it is often very weak. Transmittance mode is usually used to determine internal component concentration and internal defects of relative transparent materials such as; fruit, and vegetables. Transmittance mode has a low signal level from light attenuation and is affected by the thickness of the sample. So, to acquire images in transmittance mode, sample size should be thin that allows the light to travel through the sample.

***Interactance mode*: ** Both the light source and the detector are located in the same side of sample and parallel to each other (Fig. 1c). On the basis of such setup, the interactance mode can detect deeper information from the sample and has less surface effects compared to the reflectance mode. Interactance mode reduces the influence of thickness and this has practical advantage over the transmission mode. There is a need of special setup in the transmittance mode to seal the light in order to prevent specular reflection from directly entering to the detector.

Distinctive pattern of reflection, absorbance, transmittance and/or emitting of electromagnetic energy from different materials due to difference in their chemical composition and inherent physical structure at specific wavelength is referred as spectral signature. Hyperspectral measurements are not only surface based phenomenon. Most food products have very strong absorption of light, making them opaque over a distance of about several millimetre in visible and NIR regions. Lammertyn *et al.* (2000) calculated the light penetration depths in apple fruit. The depth was up to 4 mm in wavelength range of 700 to 900 nm. In other study, Hampton *et al.* (2003) reported that maximum penetration depth in mid-infrared region is usually a few micro meters and this is shorter than that of NIR region. The ratio of light reflected from a surface patch is often referred to as the bidirectional reflectance distribution function (BRDF) and it is a function of directions of incoming and outgoing light. The BRDF depends on the properties of the object. Material properties vary from perfect diffuse reflection in all directions (Lambertian surface) and specular reflection mirrored along the surface besides the dependency on wavelength. So, in general, diffuse reflectance is responsible for the colour of the product. More the cells involved in reflectance, the more useful will be the chemometric information obtained in the form of reflectance spectra. Basic physical aspects of light falling on surface such as tomato fruit are illustrated in Fig. 2.

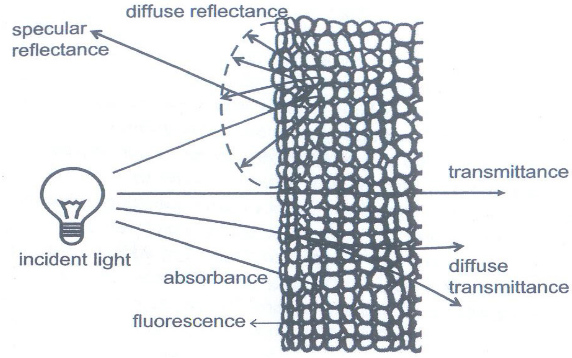

Fig. 2: Incident light on the tissue of tomato fruit results in specular reflectance, diffuse reflectance, transmittance, diffused transmittance, absorbance and fluorescence. All these physical parameters are dependent on external (surface features, colour) and internal (chemical constituents, texture) aspects of tomato fruit besides the wavelength of the light. *Source*: Polder and van der Heijden (2010).

Hyperspectral imaging/reflectance for quality analysis

In the past two decades, hyperspectral imaging has been explored intensively for analysing physical, chemical and biological properties of a broad range of food and agricultural products for quality and

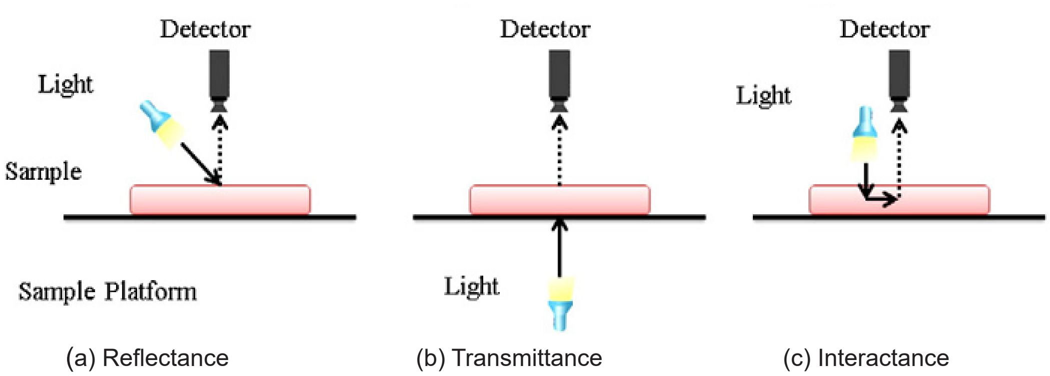

Fig. 1: Acquisition approaches of hyperspectral imaging. *Source*: Wu and Sun (2013).

safety aspects from a macroscopic approach to a more limited field area. Vis-NIR range is used to measure food quality (particularly fruit quality), determining the internal constituents of food products, online detection of diseases and chemical, microbial or biological contaminants (Gomez-Sanchis *et al*., 2012), impurity discrimination (straw, broken grains, grains from other crops, weed seeds, insects, plastic, stones, pieces of wood and paintings, animal feces) in many cereals such as wheat, spelt and barley (Fernandez Pierna *et al*., 2012; Hussain *et al*., 2019), maturity determination (Menesatti *et al*., 2008), discrimination of botanical families and plant species and also for detection of toxic and invasive plants from mixed meadows (Dale *et al*., 2011; Punalekar *et al*., 2016). Hyperspectral imaging has been used widely for evaluation of internal starch, total soluble solids, titratable acidity, water content, acidity, sugars, pH, oil content, pigments, dry matter content, stiffness, physiological defects, physiological disorders and other properties of fruits and vegetables (Magwaza *et al*., 2014; Paul *et al*., 2018).

A typical reflectance pattern of a tomato fruit at three different ripening stages is presented in **Fig. 3**. Distinct differences in the reflectance pattern are due to the ripening stages or degree of maturity of tomato fruit (Kumar *et al*., 2022a). Typical characteristics of reflectance spectra of tomato fruit as described by Sun (2010) and Szuvandzsiev *et al.* (2014) are as follows: 1) Intense absorption at 560 nm, 2) Above 560 nm, reflectance values become higher, 3) Maximum reflectance can be recorded between 645 and 713 nm, depending on the sample, 4) Green fruits show a valley near 670 nm which is due to absorption by chlorophylls, 5) With the progress of ripening chlorophyll valley disappear and an absorption valley is formed in 400-550 nm and 6) A local absorption maximum at around 980 nm is seen in near-infrared (NIR) region and this is due to the presence of carotenoids.

The hyperspectral imaging/reflectance technology has been applied in various fields related to tomato including grading, processing, and marketing etc. The power and potential of this technology can be judged from the fact that Vis-NIR diffuse reflectance spectroscopy combined with multivariate analysis was successfully used to differentiate 70 transgenic tomatoes and 94 of their parents (Xie *et al*., 2007). This was also employed for non-destructive determination of internal defect (Cho *et al*., 2013), ripeness (Polder and van der Heijden, 2010), maturity/physiological maturity and internal chemical attributes (moisture, soluble solids, pH, lycopene, β-carotene, polyphenols and firmness) of tomato fruits (Van de Poel *et al.,* 2012; Tiwari *et al*., 2013; Rahman *et al*., 2017; Huang *et al*., 2018; Alenazi *et al*., 2020; Zhao *et al*., 2022). A detailed compilation of the work done in last two decades on non-destructive assessment of different nutritional and quality parameters in tomato fruits by hyperspectral imaging/reflectance has been presented in Table 1.

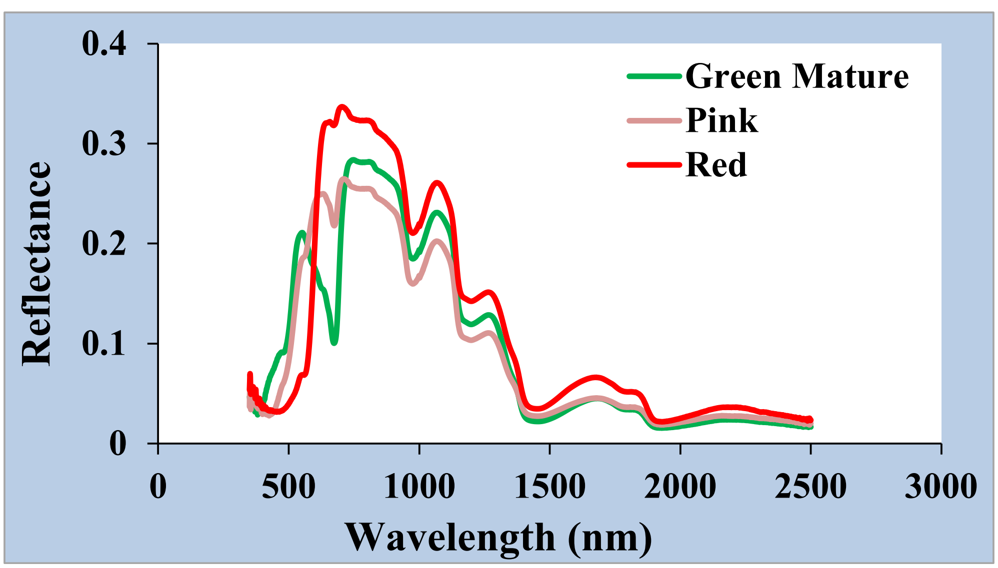

Fig. 3: Reflectance spectra of tomato fruit at three different ripening stages. *Source*: Kumar *et al.* (2022a)

Table 1: The work as done during last two decades on non-destructive assessment of different nutritional and quality parameters in tomato fruits by hyperspectral imaging/reflectance

Parameter	Wavelength range (nm)	References
Moisture content	1000-1550	Rahman *et al*. (2017)
Soluble solid content	1000-1550	Rahman *et al*. (2017)
Soluble solid content	550-1650	Huang *et al*. (2018)
Soluble solid content	650-930	Egei *et al*. (2022)
Soluble solid content	400-1000	Xiang *et al*. (2022)
pH	1000-1550	Rahman *et al*. (2017)
pH	550-1650	Huang *et al*. (2018)
pH	400-1000	Xiang *et al*. (2022)
Sweetness index	1000-1550	Rahman *et al.* (2018)
Maturity/ripeness	530, 595,630 and 850	Hahn (2002)
Maturity/ripeness	396-736	Polder *et al*. (2002)
Colour	600-1100	Kusumiyati *et al*. (2008)
Colour	430-1400	Chen (2008)
Colour (*L**, *a**, *b**, Hue, chroma)	325-985	Van Roy *et al*. (2017)
Chlorophyll *a* and *b*	450-600	Chen (2008)
Lycopene	400-600, 600-800, 800-1000, 400-1000	Pedro and Ferreira (2005)
Lycopene	450-1000	Chen (2008)
Lycopene (ripening on and off the plant)	623-1052	Kusumiyati *et al*. (2008)
Lycopene	380-950	Berra (2012)
Lycopene	550-975	Szuvandzsiev *et al*. (2014)
Lycopene	370-1040	Saad *et al*. (2014)
Lycopene	360-750, 400-1000, 450-1150	Clement *et al*. (2015)
Lycopene	950-1650	Deal *et al*. (2015)
Lycopene	500-1100	Tilahun *et al*. (2018)
Lycopene	950-1650	Ibrahim *et al*. (2018)
Lycopene	400-700	Alsina *et al*. (2019)
Lycopene	285-1200	Alenazi *et al.* (2020)
Lycopene	590-790	Saad *et al.* (2021)
Lycopene	650-930	Egei *et al*., (2022)
β-Carotene	400-600, 600-800, 800-1000, 400-1000	Pedro and Ferreira (2005)
β-Carotene	500-1100	Tilahun *et al*. (2018)
β-Carotene	950-1650	Ibrahim *et al*. (2018)
β-Carotene	400-700	Alsina et al. (2019)
β-Carotene	285-1200	Alenazi *et al.* (2020)
Polyphenols	550-975	Szuvandzsiv *et al*. (2014)
Total phenolics	285-1200	Alenazi *et al.* (2020)
Total flavonoid	285-1200	Alenazi *et al.* (2020)
Firmness	500-1100	Chen (2008)
Firmness	1100-1800	Sirisomboon *et al*. (2012)
Firmness (on and off the plant)	623-1052	Kusumiyati *et al.* (2008)
Firmness	350-2500	Ecarnot *et al*. (2013)
Firmness	716, 1000	Elsayed and Ghazy (2017)
Firmness	1000-1550	Rahman *et al.* (2018)
Firmness	285-1200	Alenazi *et al.* (2020)
Firmness	400-1000	Xiang *et al*. (2022)

Recent reports, with the use of reflectance at one or two wavelength/s, have shown that out of whole range of hyperspectral spectrum; specific wavelength/s (that too in the visible range) are also effective in prediction of various ripening and quality related parameters of tomato fruits. The indices identified and models developed were found to be valid across the fruits of tomato varieties and hybrids and equally good for harvested and stored tomato fruits as well. Index based on the reflectance value at wavelength 521 nm i.e., R_{521} yielded the best prediction model for non-destructive estimation of colour, ripeness or maturity of tomato fruit. For this, model [y (colour/ripeness/maturity score) = - 2.456 ln (x) - 1.093 where x is reflectance at 521 nm (R_{521})], with values of R^2 = 0.80, SEP = 0.87, RMSEP = 0.86 and bias = 0.09 indicated the potentiality to distinguish at least basic or standard ripening stages of tomato fruit from one another (Kumar *et al*., 2022a).

Likewise, the model for lycopene content i.e., y [µg g^{-1} fresh weight (FW)] = 0.1713x - 1.789 where x is R_{546} was identified as the best model for prediction of lycopene content up to a difference of ≥ 5.04 with biasness of 0.10 (Kumar *et al*., 2022b). In addition, the model for firmness [y (N) = 1260.800 x + 3.309] based on the index i.e., x = R_{501} (1st derivative) was the best for prediction of firmness of tomato fruits and this model can predict the firmness for a difference of ≥ 1.05 N with biasness of − 0.01 N (Kumar *et al*., 2022c). For total carotenoids in tomato fruits, model y (µg g^{-1} FW) = 1.6638 $x^{-1.353}$, wherein x is R_{582} emerged out as the best model with values of 8.78, 7.65 and - 0.12 for RMSEC, RMSEP and biasness, respectively (Kumar *et al*., 2022d). In all the above studies, developed models can be considered as simple and rapid because they are based on reflectance at single wavelength and that too in the visible range.

Advantages and disadvantages of hyperspectral imaging/reflectance

Various advantages and disadvantages of hyperspectral imaging/reflectance are as follows:

Advantages

- It is a novel technique that obtains both spatial and spectral information of an object (sample).
- It requires minimal or no sample preparation.
- It is chemical-free assessment which enables safety and environmental protection.
- It is non-invasive, so the sample could be used for time series or other purposes analysis.
- It is economical compared with traditional methods because it saves labour, time, reagent cost and other costs.
- 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 flexibility in choosing any region of interest (ROI) within 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 considered as a spectral signature and it can be saved in a spectral library.
- Hyperspectral images are reasonably independent of light source.
- Due to high spectral resolution, hyperspectral imaging provides both qualitative and quantitative measurements.
- Able to determine several constituents simultaneously in the same sample.
- Able to delineate multiple distribution of different constituents within a sample, not just the bulk composition.
- Competent in detection and discriminate of different objects even if they have similar colour, overlapped spectra or morphological characteristics.
- It allows identification of different biochemical constituents presented in a sample based on their spectral signatures because regions of similar spectral properties have similar chemical composition. This process is called as building chemical images or chemical mapping (required for constructing detailed maps of the surface composition of foods) which otherwise/ traditionally requires use of intense laboratory methods.
- Potential to detect even the diseases and defects in agricultural products and food items.
- As of today, with the use of appropriate tools available in the form of different software and statistical methods, it is possible to characterize the main sources of spectral variability and to pin point the optimum wavelength/wavebands that offer maximum information or content related information.

There are however some constrains in this hyperspectral imaging/reflectance and they are as follows:

Disadvantages

- It contains much redundant data that pose considerable challenges for data mining.
- It takes a long time for image acquisition and analysis, so pose difficulty in direct implementation in online application or automated quality evaluation.
- Slow computation speed, limitations of hardware and high cost are the major factor that limits its use. In fact, there is requirement of high speed of hardware for rapid image acquisition and analysis of huge amount of data.
- Hyperspectral data suffer from well-known problem of multicollinearity. Some multivariate analysis techniques like principal component regression (PCR) and partial least square (PLS) are often employed to overcome this problem. The effects of multicollinearity in data can only be reduced but cannot be completely removed by PCR and PLS.
- Not suitable for analysis of liquids or homogenous samples, because the value of imaging lies in the ability to visualize spatial heterogeneities in samples.
- Not suitable when the region of interest (ROI), within the surface of a sample, is smaller than a pixel or the quality attributes have no characteristic spectral absorption.
- Being indirect method, needs standardization calibration and model transfer procedures.
- Modeling and data processing is time consuming; interpretation programs are very expensive and specialists are needed for calibration and standardization.
- Potential heating effect is found in the measured hyperspectral images of food, due to presence of water.
- Cannot detect the information of constituents very deep inside the food samples.

FUTURE PERSPECTIVES

In the last two decade a great progress has been made in use of hyperspectral imaging/reflectance for the assessment of nutritional quality of tomatoes. However, still lots need to be done towards the transfer of this technique from the laboratories to the end users at small, medium and industry levels. Presently, the main limitation includes the high cost of the instrument/s. Since, most of the developed models were based on wavelength range and this increases the manufacturing and operational cost of the instrument. So, further work can also focus on assessment of nutritional and quality parameters of tomato by using a single or at the most two different wavelengths (preferably in vis-NIR region).

This will help in reducing the cost of the instrument with possibility of usage even up to the farmer's level. In the near future, identified indices and the developed models can be translated into cost-effective tools/technique with wider applicability covering not only the basic research (rapid or automated phenotyping, screening, monitoring and sorting of tomato fruits) as desired by breeders, physiologist, horticulturist, food scientists and but also the applied fields (automated colour or quality based grading of tomato fruits) as desired for better postharvest management and by marketing, processing, agri-industry value-addition and pharmaceutical sectors.

Survey of previous and recent literature indicate that future work on hyperspectral imaging/reflectance for quality assessment need to give more emphasis on the following aspects:

- To actually realize the full potential of hyperspectral imaging/reflectance as a non-destructive analytical tool for quality and physiological status determination.
- Making use of NIR region for assessment of quality parameters and plant constituents.
- Multispectral imaging should aim for acquiring the spectral images only at several optimal wavelengths to meet the speed requirement of quality assessment and inspection. Such optimized multispectral imaging systems have much lower dimensionality than the hyperspectral imaging system, resulting in taking less time in data acquisition.
- Using the above approach of multispectral imaging and thereby developing indices/models based on wave band/s or wavelength/s selection. This can give better result than the use of full or part of spectrum.
- Increasing the efficiency/robustness by identification of key/contributing wave bands or wavelengths that can give minimum error with maximum discrimination power/quantification ability.
- There is still need for improvement in interpretation of acquired data by making use/developing suitable statistical technique in the

process of chemometric analysis which is a key component of index/model development.

- Minimization of errors by taking more varieties, number of samples from different locations and over different years of harvest.

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