with our restricted focus, we refer to relevant classical papers and recent papers but also include a number of reviews containing broader and more extensive literature compilations.

6.2 Methods to determine optical properties of plant surfaces

6.2.1 UV-visible absorbance spectrophotometry

Solutions of (epi)cuticular matter obtained by short-term extraction of plant surfaces with chloroform, or aqueous/methanolic extracts of epidermal strips peeled off plant organs are often measured spectrophotometrically to assess absorption properties of plant surfaces (see Section 6.3.1). These measurements are suitable for classifying the principal absorbing molecules present, and to reveal changes in their concentration, for example, during acclimation processes. Because optical conditions within plant surfaces often differ fundamentally from those in solutions, spectrophotometric data of extracts should not be used uncritically to evaluate the absorption of these compounds *in vivo*.

Stripping off the epidermis is not possible in many plant species; consequently, whole leaf extracts have been analysed spectrophotometrically to identify and classify epidermal UV-screening compounds (for a review see Searles *et al.*, 2001). The limitations of this method obviously include not only the different optical behaviour of pigments in solutions and *in vivo*, but also the difficulty of allocating variations in concentration of UV-absorbing molecules between upper and lower epidermis and leaf mesophyll (Kolb and Pfündel, 2005).

UV-Vis spectrophotometry has also been employed to analyse mechanically isolated epidermal strips or enzymatically prepared cuticles (see Section 6.3.1). Because isolation and subsequent handling can disrupt these layers, such spectroscopic studies should be accompanied by parallel microscopic investigations to confirm the intactness of the preparations. Furthermore, these layers scatter the measuring beam so that a part of the transmitted radiation, leaving the sample at oblique angles, misses the photodetector in standard spectrophotometers; therefore, spectrophotometric measurements of scattering samples tend to underestimate the sample transmittance. It is possible to reduce this error by placing the sample close to the photodetector, thereby increasing the angle of acceptance, and also by completely diffusing all radiation transmitted by the sample with an opal quartz plate (Butler, 1964). On the other hand, fluorescence excited by the measuring radiation might result in overestimation of transmittance particularly with highly absorbing samples.

6.2.2 Integrating sphere

Integrating spheres are hollow globes with highly reflective inner surfaces. In the so-called 'external' integrating sphere, the sample forms a defined area of the inner