17.3.4 *In Vitro* Antioxidant Assays

17.3.4.1 Radical Scavenging Activity Using DPPH• Method

Relatively stable DPPH radicals have been widely used to test the ability of compounds to act as free radical scavengers or hydrogen donors and thus to evaluate antioxidant activity (Jao and Ko 2002). The reduction capacity of DPPH radicals was determined by the decrease in its absorbance at 517 nm (Duh 1998). The results of DPPH assay were expressed in IC_{50} values. The lower value of IC_{50} indicates a higher antioxidant activity. The *B. retusa* stem methanol extract (20.04 µg/mL) showed better DPPH radical scavenging activity compared to other solvent extracts, shown in Figure 17.1. From this, it is clear that methanol extract showed better radical scavenging activity by reducing the stable DPPH radicals to a yellowish diphenylpicrylhydrazine derivative. They also exhibited good antioxidant activity in the traditional food antioxidant quercetin (4.70 µg/mL) and synthetic antioxidant BHT (31.12 µg/mL), which were used as reference compounds. Similarly, Sudhanshu et al. (2012) has also been reported to have anti-DPPH· activity in *B. retusa* extracts.

17.3.4.2 Scavenging Ability on ABTS•+

The TEAC (Trolox equivalents antioxidant capacity) was measured using the improved ABTS•+ radical decolourisation assay, one of the most commonly employed methods for antioxidant capacity, which measures the ability of a compound to scavenge ABTS•+ radicals (Awika et al. 2003). The result of ABTS cation radical scavenging activity of stem extracts of *B. retusa* is shown in Table 17.2. The ethyl acetate extract of stems showed higher cation radical scavenging activity (8396.95 µM TEAC/g extract) compared to that of other solvent extracts. Hagerman et al. (1998) reported that the high molecular weight phenolics have more ability to quench free radicals (ABTS•+). This is may be due to the presence phenolic contents in the *B. retusa* extracts.

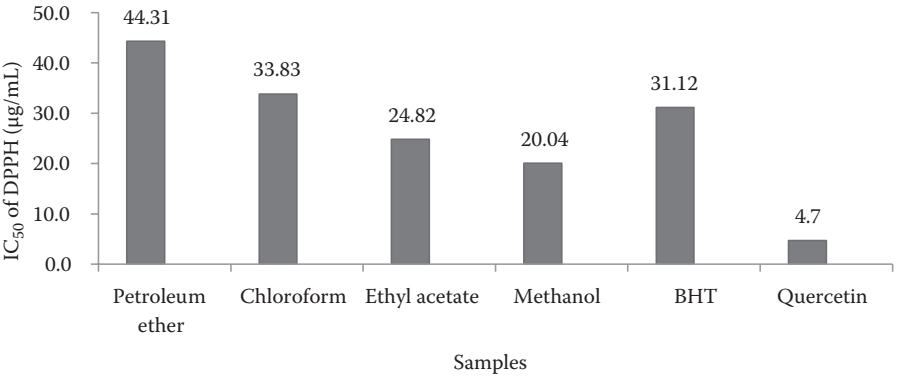

FIGURE 17.1 DPPH radical scavenging activity.