



**Figure 28** Structure of betalain pigments from *Beta vulgaris*. (A) Betacyanin: betanin (R = glucose). (B) Betaxanthins: vulgaxanthin I (R = OH) and vulgaxanthin II (R = NH<sub>2</sub>). (C) Betalamic acid.

## 2. Distribution

The occurrence of the betalains is restricted to certain plant families of the Caryophyllales (= Centrospermae) order and to toadstools, notably *Amanita*, *Hygrophorus*, and *Hygrocybe* species (62,63). These pigments may be present in flowers (cacti), roots (beetroot), fruits (*Rivinia* spp.), or bracts (*Bougainvillea* spp.). The betacyanins show a superficial similarity to the anthocyanins; however, the two groups seem to be mutually exclusive.

## B. TLC

Betalains are highly polar water-soluble compounds. They often occur as complex mixtures and are easily decomposed during the purification steps, which renders the isolation of larger amounts of pigments difficult. The most successful methods for separating these pigments have been electrophoretic techniques, column chromatography (Sephadex and Polyamide), HPLC, and TLC.

TLC on cellulose (Avicel, Macherey-Nagel) with the solvent ethyl acetate–formic acid–water (33:7:10) was used for separation of a complex mixture of betacyanins from bracts of *Bougainvillea glabra* (64). Other solvent systems for separation of betalains on cellulose include 2-propanol–ethanol–water–acetic acid (6:7:6:1) (65). When a system using silica gel as support (66) was tested in our laboratories in order to separate betalains from beetroot, it offered no advantage over the use of the above-mentioned cellulose systems. Separation of betaxanthins has been carried