

c. *Butenolide*. Butenolide has been extracted from moldy grains contaminated with the fungus *Fusarium nivale* (40). It is eluted on silica gel plates using chloroform–methanol (93:7) or toluene–ethyl acetate–formic acid (6:3:1) as solvent (41). It is also eluted on Silufol plates using chloroform–methanol (4:1), toluene–ethyl acetate–formic acid (6:3:1), or butanol–acetic acid–water (4:1:4). The separated toxin can be seen as gray spots under visible light after spraying with *p*-anisaldehyde or as yellow spots after spraying with 2,4-dinitrophenylhydrazine followed by heating at 100°C for 2–3 min (40).

d. *Patulin*. Patulin is a secondary metabolite produced by several species of molds, including *Penicillium expansus*. This mycotoxin has strong antibacterial activity against many bacteria. It is carcinogenic, mutagenic, and highly toxic to animal cells. Although apples and pears are good sources, patulin is commonly found in apple juice (43). However, it has also been found in some cereals, legumes, and sunflower seeds (44).

Silica gel is commonly used for the separation of patulin by TLC, with various solvent systems (see Table 5). After the separation on TLC plates, patulin is detected by spraying phenylhydrazinium chloride (50), 3-methyl-2-benzothiazolinone hydrazone (MBTH)–hydrochloric acid (51,52), or 4% phenylhydrazine hydrochloride followed by heating for 2–3 min (53). Patulin is clearly seen as yellow spots. Patulin in apple products is also determined by a rapid TLC scanning method using plastic-backed TLC plates precoated with silica gel containing fluorescent indicator UV<sub>254</sub> (54). The plates are developed in toluene–ethyl acetate–formic acid (6:3:1), and after drying in air the plates are sprayed with 3-methyl-2-benzothiazolinone hydrazone hydrochloride hydrate (MBTH) followed by heating at 130°C for 15 min. Fluorescent spots are measured by scanning densitometry.

e. *Citreoviridin*. Citreoviridin mycotoxin is produced by *P. charlesii* and is extracted from mold-contaminated rice (55) and pecans (56). It is separated on silica gel TLC plates with chloroform–methanol–dimethyl ketone (45:3:2), chloroform–methanol (9:1), or ethyl acetate–toluene (1:1) (57). Solvent systems commonly used for the separation of other mycotoxins, such as ethyl acetate–toluene (1:1), chloroform–methanol (9:1), and chloroform–methanol–dimethyl ketone (45:3:2) (57), are also used for the separation of citreoviridin on silica gel plates. Ethyl acetate–toluene (3:1) is used on Kiesel-gel G 1500 (58), whereas Silufol plates are developed in benzene–

**Table 5** Solvent Systems for the Separation of Patulin by TLC

Adsorbent	Solvent system <sup>a</sup>	Ref.
Silica gel	Methanol (100%), C:M (1:1), chloroform (100%), E:W (4:1), T:EA:FA (6:3:1), B:P:W (2:2:1), B:M:AA (24:2:1)	45, 46
Silica gel 60	C:DMK (90:10), C:M (95:5), T:AA:FA (90%) (50:40:10), Pen:EA (96:4), DIPE:P:E:Pyr (84:12:4:0.8)	39
Silica gel F <sub>254</sub>	B:C:DMK (45:40:15), C:M (97:3), C:DMK:P (85:15:20), C:DMK:H (7:2:1), C:DMK (9:1)	26
Silica gel G-HR	T:EA:FA (5:4:1)	46
Silical gel K-5	T:EA:FA (95%) (5:4:1)	47
Kiesel-gel 60 F	DCM:EA (95:45)	48
Kiesel-gel 60 G	T:EA:FA (85%) (50:40:10)	49
Silufol	B:M:AA (24:2:1), T:EA:FA (90%) (6:3:1), B:E (95:5), C:M (4:1), C:MIBK (4:1), C:DMK (9:1), C:AA:DEE (17:1:3), But:AA:W (4:1:4)	27

<sup>a</sup>B = benzene, But = butanol, C = chloroform, DCM = dichloromethane, DEE = diethyl ether, DIPE = diisopropyl ether, DMK = dimethyl ketone, E = ethanol, EA = ethyl acetate, FA = formic acid, H = hexane, M = methanol, MIBK = methyl isobutyl ketone, P = propanol, Pen = pentane, Pyr = pyridine, T = toluene, W = water.