Restriction analysis of plasmids

- Solutions: - 1x TBE: make from 10xTBE

10xTBE:	0.89M	Tris base	109 g
	0.89M	H_3BO_3	55 g
	0.025M	EDTA	9.3 g
			add H_2O to 1l

- 10x loading buffer:

20%	Ficoll 400		10 g
	0.1 M	EDTA	10 ml 0.5 M
	1%	SDS	5 ml 10%
	0.25%	bromphenolblue	0.125 g
	0.25%	xylencyanol	0.125 g
			add H_2O to 50 ml

- use ~200 ng plasmid DNA
- 10 µl sample:
  - 1 µl 10x buffer
  - x µl DNA
  - x µl restriction enzyme (1 U cuts 1 µg DNA in 1 h at 37°C)
  - (0.1 µl BSA 10 mg/ml if needed)
  - add H_2O to 10 µl
- incubate 1 h at 37°C
- add 1 µl 10x loading buffer
- 0.8% agarose gel:
  - dissolve 0.8 g agarose in 100 ml TBE-buffer (heat in microwave)
  - add 5 µl 10 mg/ml ethidium bromide (= 0.5 µg/ml)
    (wear gloves, EtBr is carcinogenic!)
  - pour into gel box and let polymerize (~45 min)
- load samples on gel and run at 5-10 V/cm length of gel
- don't forget to add 5 µl of DNA ladder as standard
- when blue markers are sufficiently separated, take a photograph on UV table