Fermentas GeneJET Gel Extraction

- Excise desired band from agarose gel, remove as much agarose as possible. Cut gel slice into halves or quarters, put gel pieces into an Eppendorf tube
- Weigh gel slice on analytical balance.
- Add 2X gel-weight of Binding Buffer to the excised gel piece (for example, if gel slice weighs 250 mg, add 500 uL of Binding Buffer)
- Incubate at 55°C until agarose dissolves (about 10-15 minutes)
- Vortex the solution, ensure that all agarose has dissolved
- **IF YOUR DNA FRAGMENT IS ≤500bp or ≥10,000bp, ADD 1X GEL-WEIGHT OF ISOPROPANOL TO THE DISSOLVED AGAROSE SOLUTION AND VORTEX**
- Add up to 750 uL of dissolved agarose solution to spin column
- Centrifuge at 14,000 rpm for 30 seconds
- Pour the flow-through from the collection tube, reinsert column
- Add 100 uL of Binding Buffer to the column
- Centrifuge at 14,000 rpm for 30 seconds
- Pour the flow-through from the collection tube, reinsert column
- Add 700 ul of Wash Buffer to the column
- Centrifuge at 14,000 rpm for 30 seconds
- Pour the flow-through from the collection tube, reinsert column
- Centrifuge at 14,000 rpm for 1 minute to dry the column
- Discard the collection tube, place column in clean Eppendorf tube
- Add 50 uL of Elution Buffer directly to the resin (can use 30 uL of Elution Buffer in order to concentrate the DNA)
- Incubate the column at room temperature for 2 minutes
- Centrifuge at 14,000 rpm for 1 minute
- Discard column