



## QIAGEN Supplementary Protocol:

### Scalable purification of archive-quality DNA from 100 to 5 x 10<sup>8</sup> cultured cells using the Gentra<sup>®</sup> Puregene<sup>®</sup> Cell Kit

This protocol provides information about scaling of reagents required for purification of DNA from 100 to 5 x 10<sup>8</sup> cultured cells using the Gentra Puregene Cell Kit.

The Gentra Puregene Cell Kit enables convenient, scalable purification of DNA from cultured cells. Reagent volumes are scaled proportionately according to the amount of starting material. Tables 1–4 show the volumes of reagents required for DNA purification from 100 to 5 x 10<sup>8</sup> cultured cells. The information provided in Tables 1–4 is intended to supplement the information given in “Protocol: DNA Purification from Cultured Cells Using the Gentra Puregene Cell Kit” in the *Gentra Puregene Handbook*.

**IMPORTANT:** Please read the *Gentra Puregene Handbook*, paying careful attention to the safety information, before beginning this procedure. For safety information on the additional chemicals mentioned in this protocol, consult the appropriate material safety data sheets (MSDSs), available from the product supplier. The Gentra Puregene Cell Kit is intended for molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of a disease.

**Table 1. Purification of DNA from 100 – 1 x 10<sup>6</sup> cultured cells**

	Number of cells		
	100–10,000	10,000-100,000	0.5–1 x 10 <sup>6</sup>
<b>Tube size (ml)</b>	0.6	1.5	1.5
<b>Volume of Cell Lysis Solution (ml)</b>	0.06	0.1	0.15
<b>Volume of RNase A Solution (μl)</b>	0.5	0.5	0.75
<b>Volume of Protein Precipitation Solution (ml)</b>	0.02	0.033	0.05
<b>Volume of 100% isopropanol (ml)</b>	0.06	0.1	0.15
<b>Volume of 70% ethanol (ml)</b>	0.06	0.1	0.15
<b>Volume of DNA Hydration Solution (μl)</b>	10	10	25
<b>Typical DNA yield (μg)</b>	0.0004–0.04	0.04–0.4	2–6

**Table 2. Purification of DNA from 1–9 x 10<sup>6</sup> cultured cells**

	Number of cells		
	1–2 x 10 <sup>6</sup>	3–5 x 10 <sup>6</sup>	6–9 x 10 <sup>6</sup>
Tube size (ml)	1.5	1.5	2
Volume of Cell Lysis Solution (ml)	0.30	0.60	0.75
Volume of RNase A Solution (μl)	1.5	3	4.5
Volume of Protein Precipitation Solution (ml)	0.1	0.2	0.25
Volume of 100% isopropanol (ml)	0.3	0.6	0.75
Volume of 70% ethanol (ml)	0.3	0.6	0.75
Volume of DNA Hydration Solution (μl)	50	100	100
Typical DNA yield (μg)	5–10	15–30	30–45

**Table 3. Purification of DNA from 1–9 x 10<sup>7</sup> cultured cells**

	Number of cells		
	1 x 10 <sup>7</sup>	5 x 10 <sup>7</sup>	6–9 x 10 <sup>7</sup>
Tube size (ml)	15	15	50
Volume of Cell Lysis Solution (ml)	1.5	6	10
Volume of RNase A Solution (μl)	7.5	30	50
Volume of Protein Precipitation Solution (ml)	0.5	2	3.3
Volume of isopropanol (ml)	1.5	6	10
Volume of 70% ethanol (ml)	1.5	6	10
Volume of DNA Hydration Solution (μl)	125	500	750
Typical DNA yield (μg)	40–60	150–300	280–430

**Table 4. Purification of DNA from 1–5 x 10<sup>8</sup> cultured cells**

	Number of cells		
	1 x 10 <sup>8</sup>	2 x 10 <sup>8</sup>	5 x 10 <sup>8</sup>
<b>Tube size (ml)</b>	50	250	250
<b>Volume of Cell Lysis Solution (ml)</b>	15	30	75
<b>Volume of RNase A Solution (μl)</b>	75	150	375
<b>Volume of Protein Precipitation Solution (ml)</b>	5	10	25
<b>Volume of isopropanol (ml)</b>	15	30	75
<b>Volume of 70% ethanol (ml)</b>	15	30	75
<b>Volume of DNA Hydration Solution (μl)</b>	1000	2000	2000
<b>Typical DNA yield (μg)</b>	200–600	400–1200	1000–3000

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