

## 830: R2A MEDIUM

This recipe contains strain-specific modifications for *Nocardioides ginsengagri* DSM 21362 \*

Final pH: 7.2

Final volume: 1000 ml

Yeast extract	0.50	g
Proteose peptone (Difco no. 3)	0.50	g
Casamino acids	0.50	g
Glucose	0.50	g
Starch (soluble)	0.50	g
Na-pyruvate	0.30	g
K <sub>2</sub> HPO <sub>4</sub>	0.30	g
MgSO <sub>4</sub> x 7 H <sub>2</sub> O	0.05	g
Agar	15.00	g
<b>Modified Wolin's mineral solution</b>	<b>1.00</b>	<b>ml</b>
<b>Trace element solution SL-10</b>	<b>0.01</b>	<b>ml</b>
Distilled water	1000.00	ml

Final pH 7.2; adjust with crystalline K<sub>2</sub>HPO<sub>4</sub> or KH<sub>2</sub>PO<sub>4</sub> before adding agar. Add agar, heat medium to boiling to dissolve agar, and autoclave for 15 min at 121°C.

\* 1 drop of 10x Wolin Vitamine Solution (see medium 141) and 10 microliter trace element solution SL-10 (see medium 320) for better growth in liquid

### **Modified Wolin's mineral solution\*** (from medium 141)

Nitrilotriacetic acid	1.50	g
MgSO <sub>4</sub> x 7 H <sub>2</sub> O	3.00	g
MnSO <sub>4</sub> x H <sub>2</sub> O	0.50	g
NaCl	1.00	g
FeSO <sub>4</sub> x 7 H <sub>2</sub> O	0.10	g
CoSO <sub>4</sub> x 7 H <sub>2</sub> O	0.18	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.10	g
ZnSO <sub>4</sub> x 7 H <sub>2</sub> O	0.18	g
CuSO <sub>4</sub> x 5 H <sub>2</sub> O	0.01	g
AlK(SO <sub>4</sub> ) <sub>2</sub> x 12 H <sub>2</sub> O	0.02	g
H <sub>3</sub> BO <sub>3</sub>	0.01	g
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	0.01	g
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	0.03	g
Na <sub>2</sub> SeO <sub>3</sub> x 5 H <sub>2</sub> O	0.30	mg
Na <sub>2</sub> WO <sub>4</sub> x 2 H <sub>2</sub> O	0.40	mg
Distilled water	1000.00	ml

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First dissolve nitrilotriacetic acid and adjust pH to 6.5 with KOH, then add minerals. Adjust final to pH 7.0 with KOH.

### Trace element solution SL-10\* (from medium 320)

HCl (25%)	10.00	ml
FeCl <sub>2</sub> x 4 H <sub>2</sub> O	1.50	g
ZnCl <sub>2</sub>	70.00	mg
MnCl <sub>2</sub> x 4 H <sub>2</sub> O	100.00	mg
H <sub>3</sub> BO <sub>3</sub>	6.00	mg
CoCl <sub>2</sub> x 6 H <sub>2</sub> O	190.00	mg
CuCl <sub>2</sub> x 2 H <sub>2</sub> O	2.00	mg
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	24.00	mg
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	36.00	mg
Distilled water	990.00	ml

First dissolve FeCl<sub>2</sub> in the HCl, then dilute in water, add and dissolve the other salts. Finally make up to 1000.00 ml.