Supplementary information for "Characterization and application of Cyclic β-(1, 2)-glucan produced from *R. meliloti*"

The column chromatogram of purified glucan (Fig. S1), TLC (S2), ^{1}H NMR (S3) 2D NMR (S4-S6), DSC spectrum (S7), TGA (S8), Cytotoxicity (S8), NMR spectrum of drug encapsulated glucan (S9), Particle size (S10), Cytotoxicity of glucan (S11), Antioxidant activity (S12) structure of the drugs and dyes used for glucan encapsulation (S13), and Chemical shifts of glucan (Table S1), ^{1}H NMR and FTIR spectrum of drug carrier complex (Table S2)

Fig. S1. Amount of cyclic β-(1, 2)-glucan in various fractions eluted from the size exclusion chromatography with 5% acetic acid. Fractions of 1ml were collected and assayed for carbohydrate. The amount of carbohydrate in the 33rd fraction was 8.88 mg/ml.

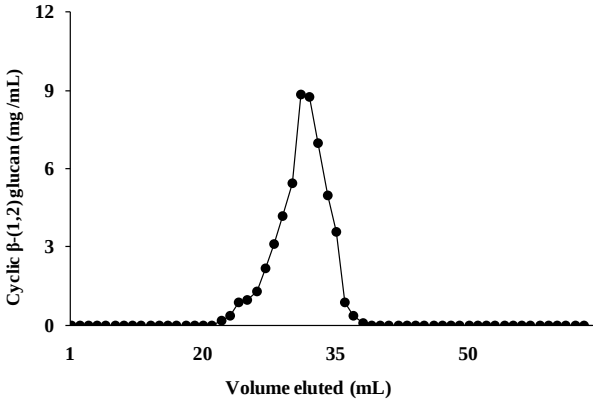


Fig. S2. Thin layer chromatography of the 33rd fraction showing the presence cyclic β-(1, 2)-glucan

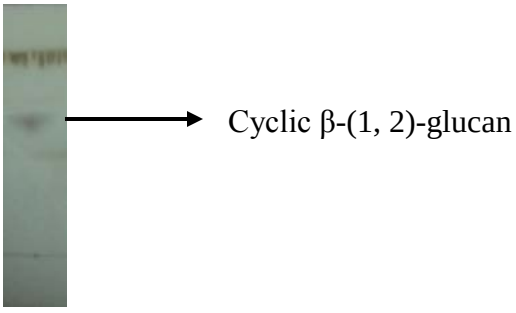


Fig. S3 ^{1}H NMR spectrum of cyclic β-(1, 2)-glucan

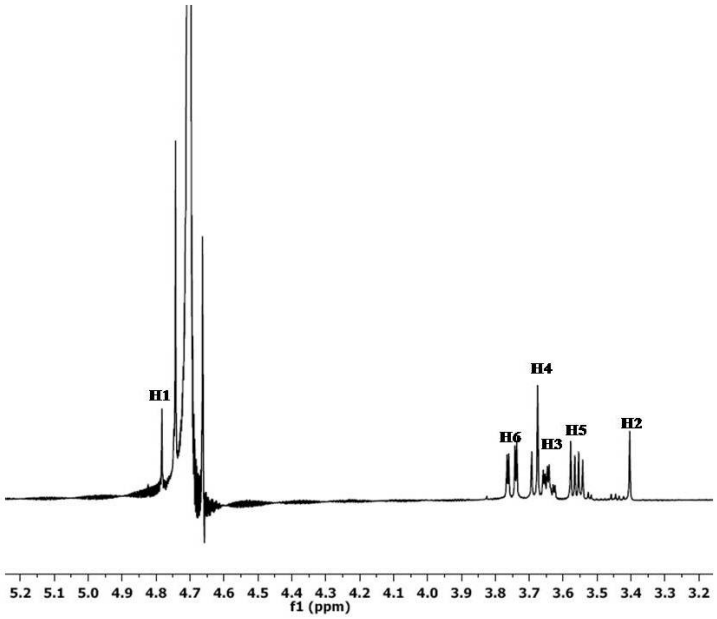


Fig. S4. ^{1}H ^{1}H NMR spectrum of cyclic β-(1,2)-glucan

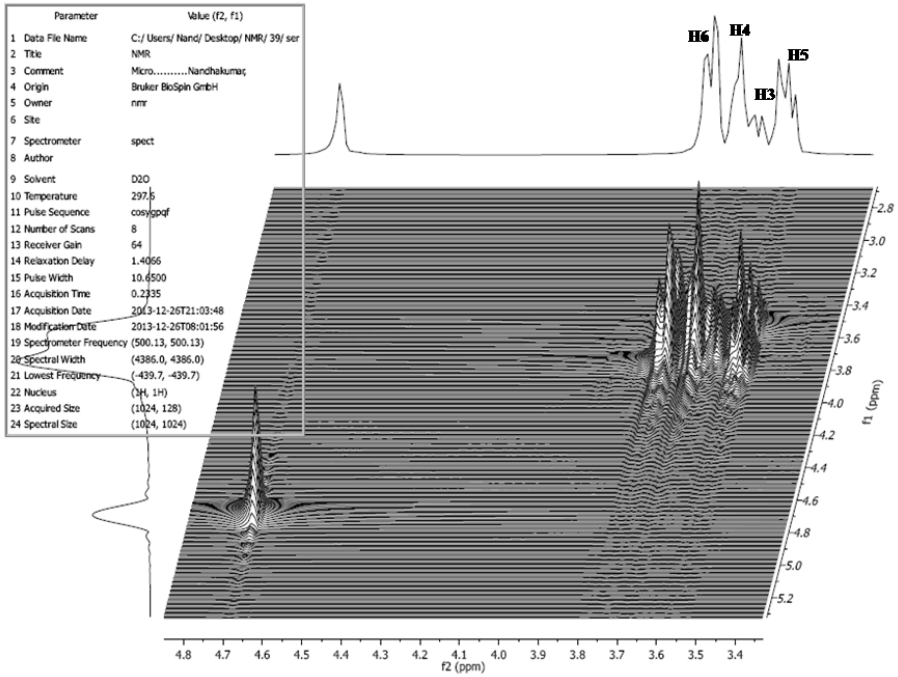


Fig. S5. HSQC spectrum of cyclic β-(1, 2)-glucan

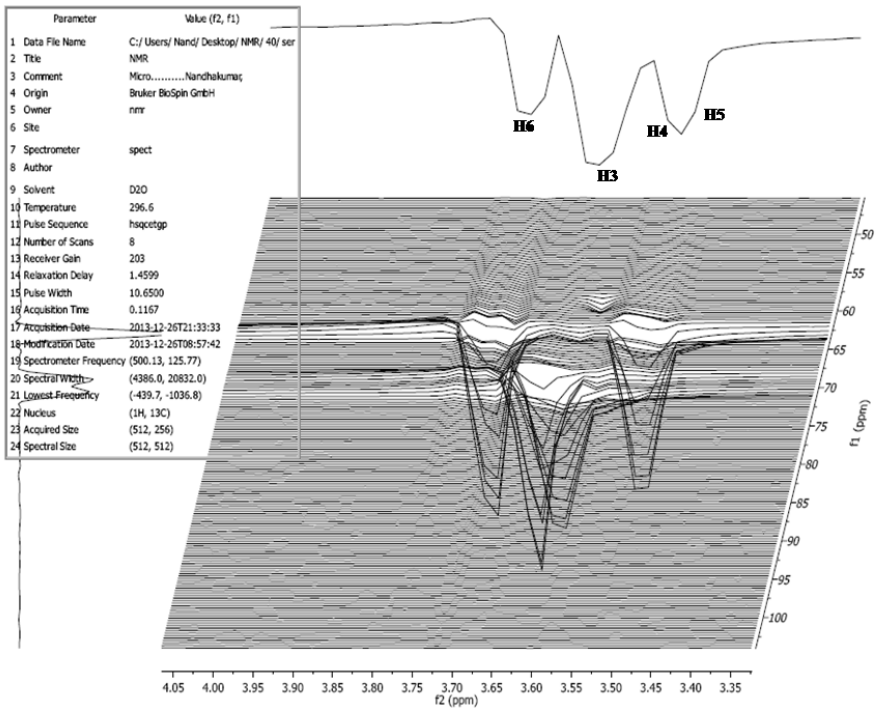


Fig. S6. HMBC spectrum of cyclic β-(1, 2)-glucan

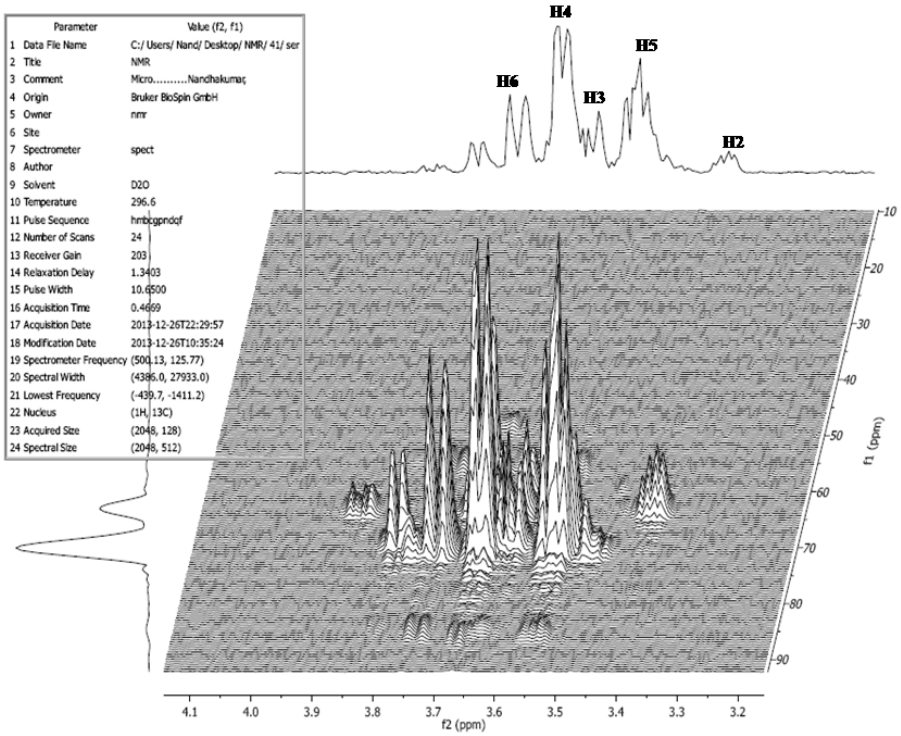


Fig. S7. Differential scanning calorimetry of cyclic β-(1, 2) glucan.

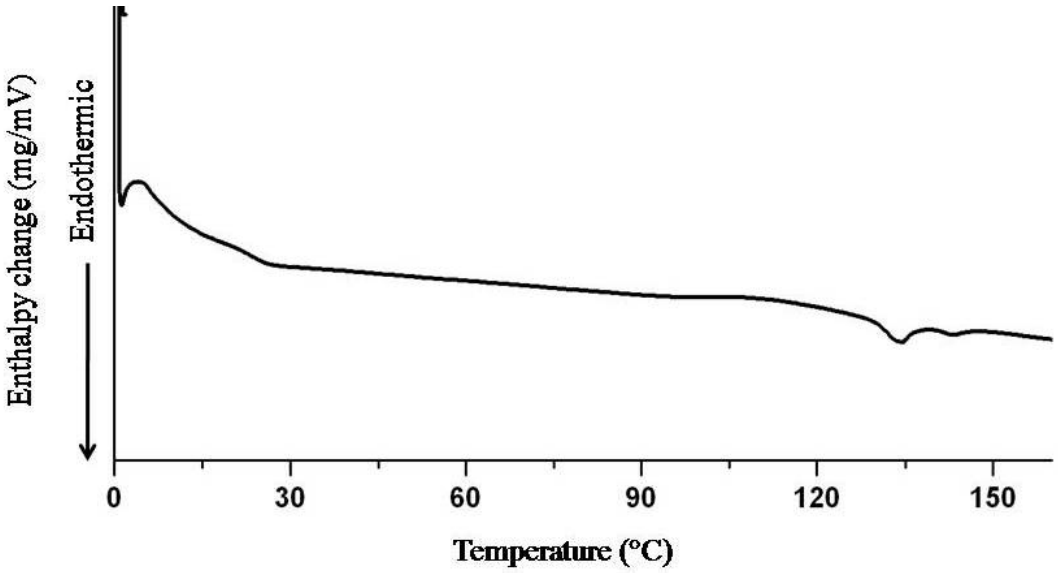


Fig. S8. Thermal gravimetric spectrum of cyclic β-(1, 2) glucan.

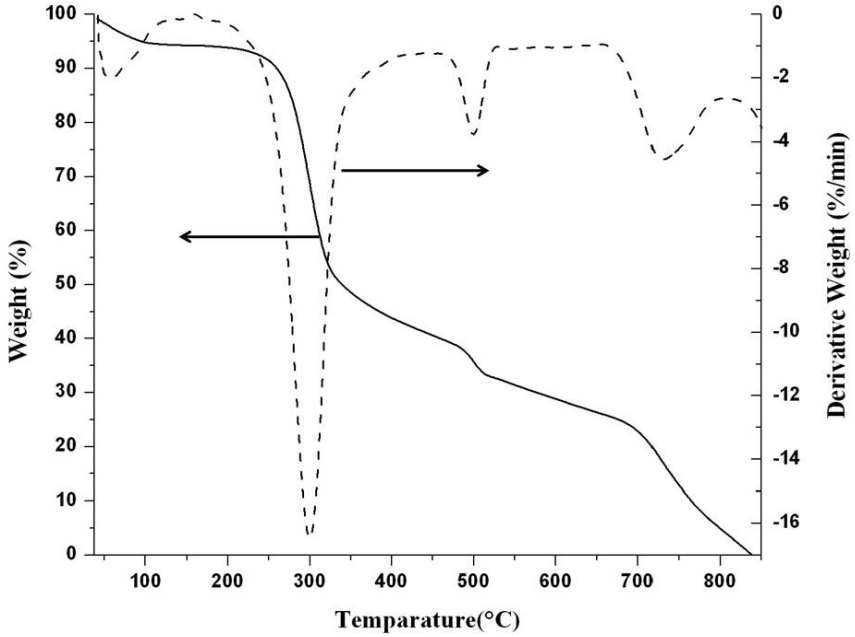


Fig. S9. ^{1}H NMR spectrum of drug encapsulated cyclic β-(1, 2)-glucan. (A) Curcumin (B) Dexamethasone (C) Reserpine (D) 6-methylcoumarin (E) 4-Hydroxy coumarin (F) 4-methyl umbelliferone.

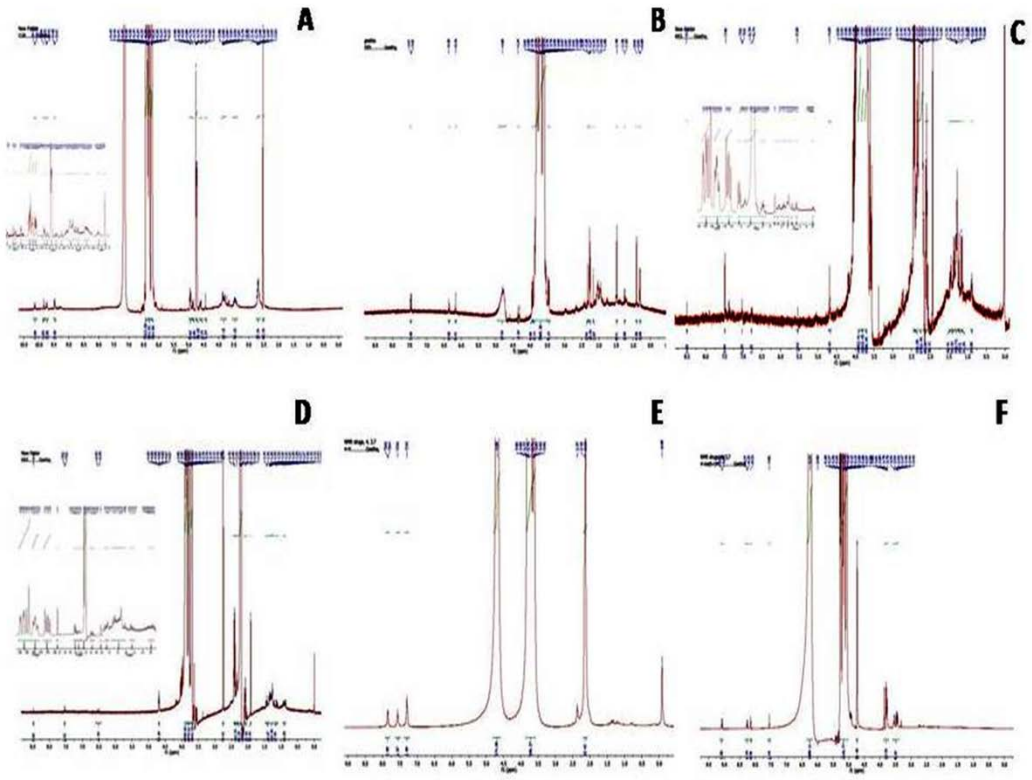


Fig. S10. Particle size distribution of curcumin encapsulated cyclic β-(1, 2)-glucan

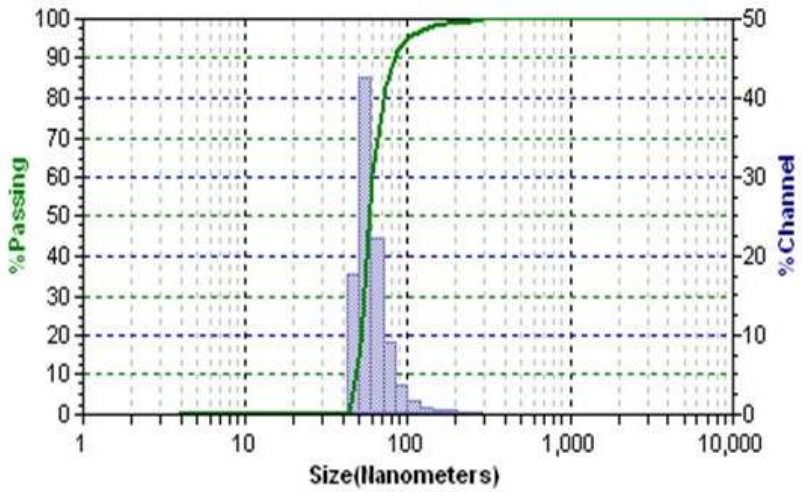


Fig. S11. Cytotoxicity potential of cyclic β-(1, 2)-glucan at different concentrations against L6 cell lines

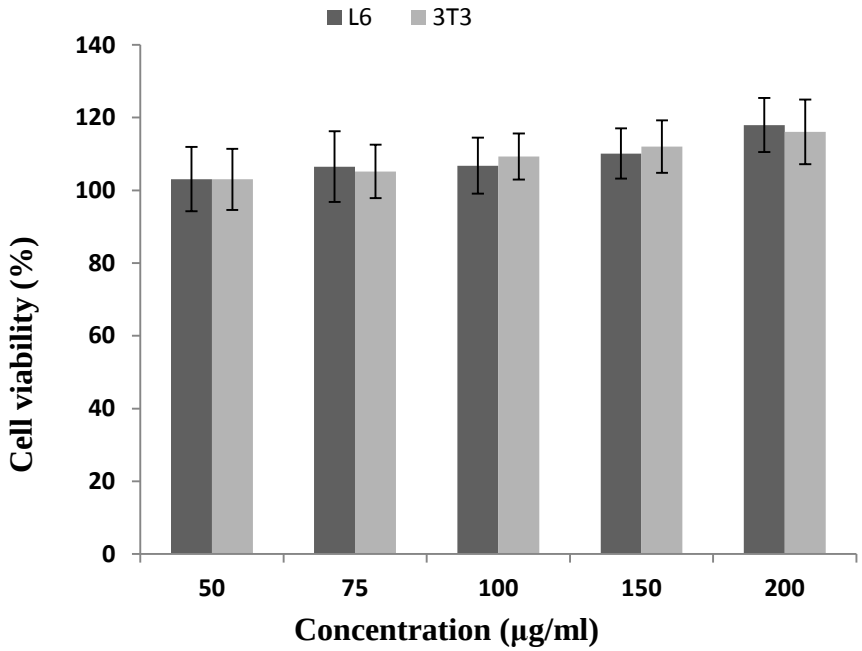
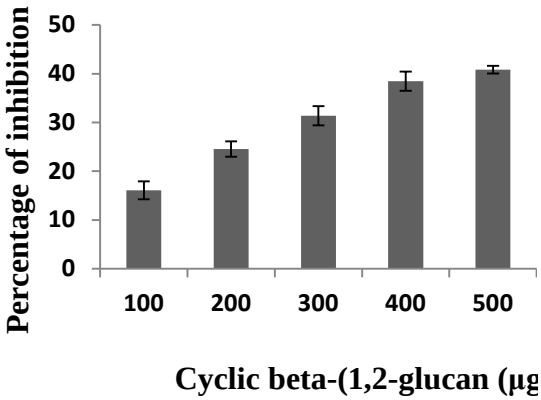


Fig. S12. Antioxidant activity of cyclic β-(1, 2)-glucan at different concentrations

a) DPPH assay



b) Hydroxyl radical scavenging assay

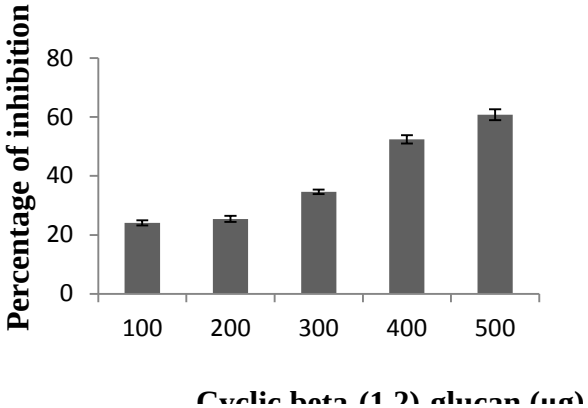


Fig. S13. Structure of different hydrophobic drugs (a) and dyes (b) used for cyclic β-(1, 2)-glucan application

a. **b.**

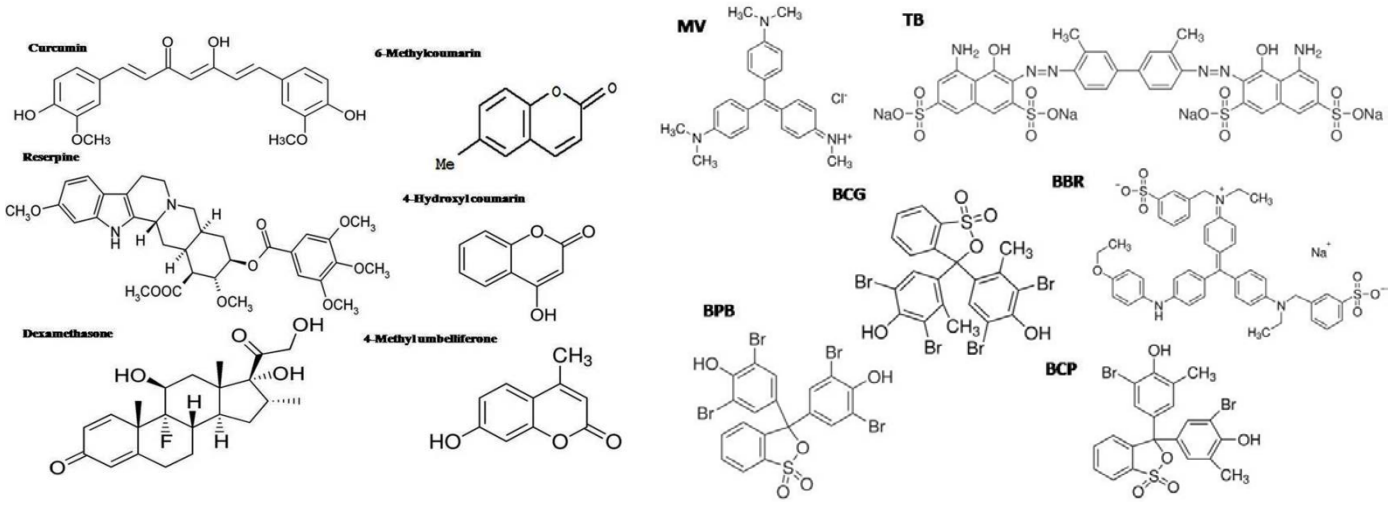


Table S1. ^{1}H chemical shifts of cyclic β-(1, 2) glucan in D_2O

Position of Protons	δ_H [Chemical shifts (ppm)[a]]
H-1	4.74 - 4.78 (m)
H-2	3.44(d)
H-3	3.60(q)
H-4	3.72 (s) ,3.70 (s) , 3.40 (s)
H-5	3.57
H-6	3.76-3.78 (dd J1=5Hz, J2=5Hz)

a Multiplicity- s-singlet; d-doublet; t-triplet, br-broad (singlet)

Table S2. ^{1}H NMR and FTIR spectrum of drug encapsulated cyclic β-(1, 2)-glucan

Name of the polymer	^{1}H NMR spectrum	FTIR spectrum
Cyclic β-(1, 2)-glucan	δ 4.75 (s, CH2), 3.81(d, J = 5Hz, CH2), 3.79 (d, J = 5Hz, OH), 3.74 (m, OH), 3.69 (m, CH), 3.62 (m, CH) ppm	3600 and 3200 cm^{-1} - O-H stretching and hydrogen-bonded hydroxyl groups. 1,000–1,100 cm^{-1} - Glycosidic bond.
Curcumin encapsulated glucan	*δ* 10.15 (d J=20Hz), 9.84 (s), 9.74 (d J=5Hz), 9.48 (d J=5Hz), 6.43 – 6.38 (m), 6.34 -6.31 (m), 6.26 (q J=10Hz), 4.90 (t J= 10Hz), 4.87 -4.85 (m), 4.76 -4.73 (m), 4.63 -4.59 (m), 4.44 (s), 3.95 -3.66 (m), 3.48 -3.40 (m), 2.72 -2.64 (m), 2.52 (s).	Aliphatic C=C (1602), Aromatic C=C (1508), keto Carbonyl (1627), C-O Stretch (1282) hydroxyl (3498), C-O-C Stretch (asymmetric 1232, symmetry 1027), aromatic CH Stretch (3013), aliphatic CH Stretch (3000), aromatic methyl CH (2945).
Dexamethasone encapsulated glucan	*δ* 7.47 (d, J=10Hz, 1H), 6.36 (d, J=5Hz, 1H), 6.16 (s, 1H), 4.80 (s,17H), 4.34 (s,3H), 3.81-3.58(m), 2.33-2.25 (m), 2.16(s,2H), 1.48 (s,7H), 1.26 (d, J=5Hz, 3H), 0.91(s,5H), 0.81 (d, J=5Hz,5H).	Hydroxyl (3471), cyclic keto carbonyl (1703), acid carbonyl (1662), C=C (1618), C-F Stretch (1135)
Reserpine encapsulated glucan	*δ* 8.50 (s), 7.46 (s), 8.02 (d, J=5Hz), 6.79 (d, J= 25Hz), 5.56 (s), 4.69 (s), 3.39-3.86 (m). 3.83 -3.79 (m), 3.74 (q, J=5Hz), 2.42 (t J=5Hz), 2.36 -2.32 (m), 2.25 -2.21 (m), 2.13 -2.05 (m), 1.93 (s), 1.4 -1.35 (m), 1.31 -1.27 (m), 1.23 (s), 1.17 (d J=5Hz), 0.90-0.88 (m).	C=C Stretch (1625), carbonyl (1730, 1710), C-O Stretch (1273), N-H Stretch (3433), C-N Stretch (1331), C-O-C Stretch (symmetry 1062, asymmetry 1225), aromatic CH Stretch (2984, 2937).
6-methylcoumarin encapsulated glucan	*δ* 8.42 (s), 7.53 (d J=5Hz), 6.55 (d J=10Hz), 4.69-4.67 (m), 3.90 -3.84 (m), 3.81 (ddd, J= 5Hz), 3.72 (q J=5Hz), 2.74 (s), 2.42 – 2.32 (m), 2.24 -2.20 (m), 2.11 -2.03 (m), 1.91 (s), 1.46-1.42 (m), 1.35 -1.21 (m), 1.15 -1.10 (m), 0.93 -0.85 (m).	C=C Stretch (1685), cyclic keto Carbonyl (1720), C-O Stretch (1262), Aromatic CH Stretch (3080, 2921)
4-hydroxy coumarin encapsulated glucan	7.86 (d, J= 10Hz, 1H), 7.56 (s, 1H), 7.30 (S, 2H), 4.69 (s),	Hydroxyl group (3182), CH stretching (aromatic) (2930),

	3.82 -3.62 (m), 2.37 -2.13 (m).	carbonyl (1665-), C=C (1606), C-O (1301)
4-methyl umbelliferone encapsulated glucan	9.08 (d, J= 5Hz, 1H), 8.27 (d, J= 10Hz, 1H), 8.15 (s, 1H), 7.55 (s, 1H), 6.24 (s), 5.28-5.06 (m), 3.86-3.76 (m), 3.54 -3.43 (m).	Hydroxyl (3185), aromatic and aliphatic CH stretching (2930, 2854) Carbonyl (1736), C=C (1664), C-O stretching (1249)