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LAMPIRAN

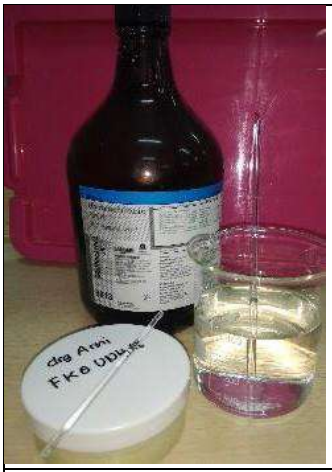
1. PEMBUATAN DDM

		
1	**2**	**3**
		
4	**5**	

Keterangan Gambar Pembuatan DDM

1. Pengumpulan limbah Gigi
2. Pembersihan karang gigi yang menempel
3. Pemisahan mahkota gigi dan akar gigi
4. Proses perendaman akar gigi
5. Peroses demineralisasi

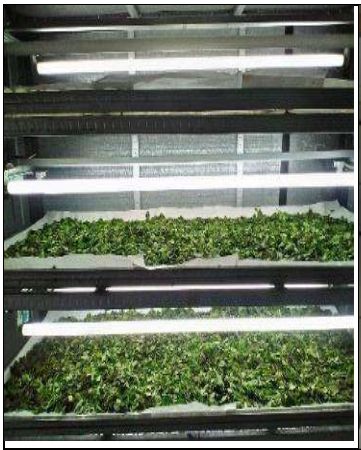
2. TAHAP DEMINERALISASI & FREEZE DRIED

		
1	2	3
		
4	5	6

Keterangan Gambar Tahap Demineralisasi

1. Persiapan alat dan bahan
2. Perendaman serbuk dentin dengan
3. Serbuk dentin yang telah ditiriskan
4. Freeze Dentin
5. Alat untuk dried
6. Hasil denti yang telah di freeze dried

3. TAHAP PEMBUATAN EKSTRAK DAUN KELOR



1

2

3

4

5

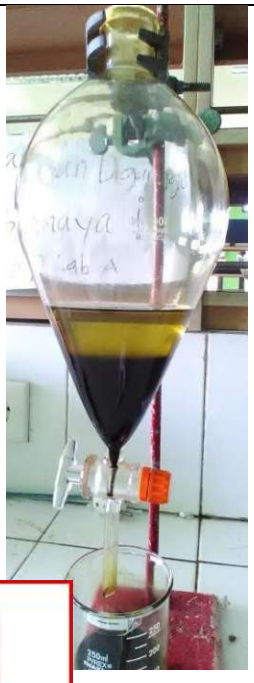
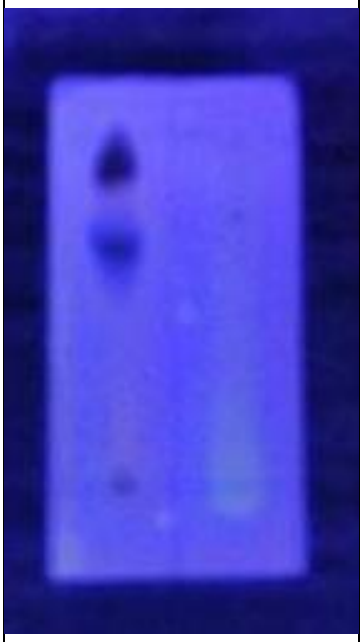
6

7

Keterangan Gambar Tahap Pembuatan ekstrak daun kelor

1. Tahap pengeringan
2. Penimbangan kelor yang telah dikeringkan
3. Perendaman dengan larutan etanol 70)
4. Proses penyaringan
5. Hasil Penyaringan
6. Proses evaporasi
7. Proses menarik air
8. Hasil ekstrak kental
9. Hasil evaporator
10. Hasil menarik air

4. Tahap Partisi

		
1	2	3
		
4	5	6

5. Tahap Freeze Dried Kelor

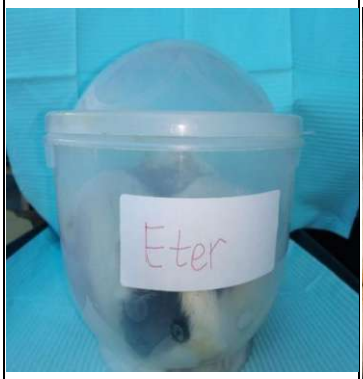
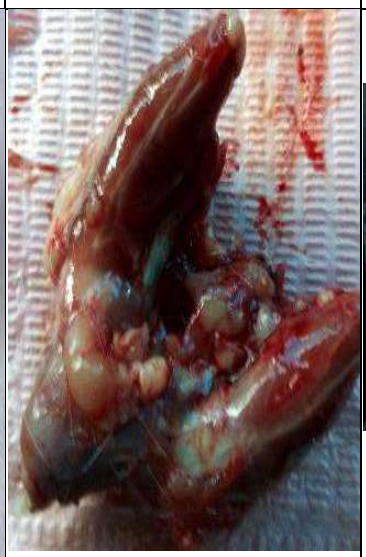
		
1	2	3

6. Tahap Pembuatan Salep

		
1	2	3
		
4	5	6
		
7	8	9

SEDIAAN SALEP

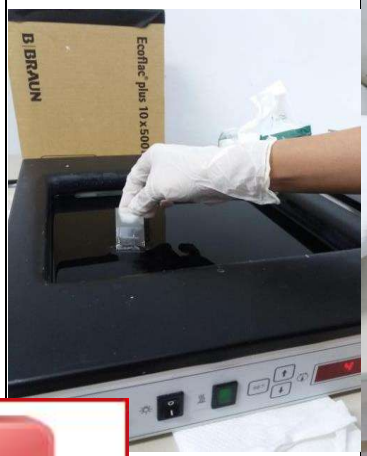
7. Aplikasi Salep ke Hewan Coba

		
1	2	3
		
4	5	6
		
7	8	

PROSEDUR PEMBUATAN SEDIAAN JARINGAN HISTOPATOLOGI DAN KETERANGAN GAMBAR

1. Jaringan yang telah dipotong dimasukkan ke dalam kaset dan diproses di dalam mesin prosesing jaringan (Tissue Automatics Prosessor).(1)
2. Proses Embedding (jaringan yang telah diproses dalam mesin prosesingan diblok menggunakan parafin cair).(2,3,4)
3. Potong jaringan dalam blok paraffin menggunakan mikrotom dengan ketebalan 3-4µm.(5)
4. Pita jaringan yang terbentuk dicelupkan ke dalam Waterbath.(6)
5. Ambil potongan jaringan dengan slide lalu tiriskan.(7)
6. Tuliskan kode pada slide sesuai dengan kode yang tertera pada blok paraffin menggunakan pensil.(8)
7. Panaskan slide diatas Hot Plate selama 1 jam.(9)
8. Dinginkan slide lalu masukkan kedalam keranjang slide. (9)
9. Deparafinasi (Xylol I, Xylol II, Xylol III) masing-masing 5 menit (10)
10. Rehidrasi (Alkohol 96%, Alkohol 80%, Alkohol 70%), masing-masing selama 5 menit.(11)
11. Cuci air mengalir selama 5 menit (12)
12. Rendam dengan hematoxylin Meyer 7-10 menit (13)
13. Cuci air mengalir selama 5 menit (14)
14. Celup-celup kedalam larutan Eosin 10 detik (14)
15. Dehidrasi (Alkohol 70%, Alkohol 80%, Alkohol 96%) masing-masing 5 menit (15)
16. Clearing (Xylol I, Xylol II, Xylol III) masing-masing 5 menit.(16)
17. Keringkan slide lalu tetesi dengan entelan dan tutup dengan deck glass. (16)
18. Amati di Mikroskop. (16)

8. Prosedur Histologi

		
1	2	3
		
4	5	6
		
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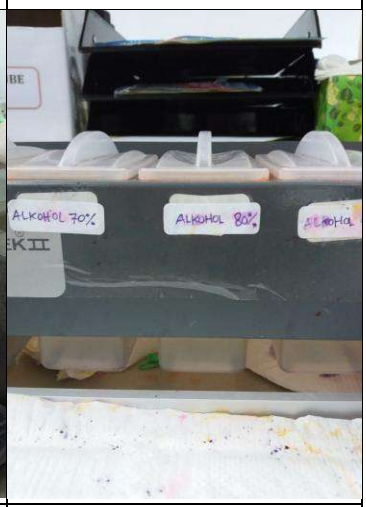

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PROSEDUR PEMBUATAN SEDIAAN JARINGAN IMMUNOHISTOKIMIA DAN KETERANGAN GAMBAR

1. Potong blok paraffin dengan mikrotom pada ketebalan 3-4µ (1)
2. Celupkan kedalam Waterbath (2)
3. Ambil potongan jaringan dengan slide lalu tiriskan (3)
4. Tulis pada slide kode sesuai blok paraffin dengan pensil (4)
5. Panaskan slide diatas Hot Plate selama 1 jam (5)
6. Dinginkan slide lalu masukkan kedalam keranjang slide (5)
7. Deparafinasi (Xylol I, Xylol II, Xylol III) masing-masing 5 menit (6)
8. Rehidrasi (Alkohol 96%, Alkohol 80%, Alkohol 70%), masing-masing selama 5 menit (7)
9. Cuci air mengalir selama 5 menit (8)
10. Angkat dari air lalu masukkan slide kedalam larutan Blocking Endogen Peroksida, rendam selama 15 menit (9)
11. Cuci air mengalir selama 5 menit (10)
12. Masukkan keranjang berisi slide kedalam decloaking yang berisi larutan Antigen Retrival Decloaking Chamber, lalu letakkan slide pada rack holder(11)
13. Masukkan rack holder kedalam decloaking, lalu tutup (11)
14. Atur waktu yaitu selama 40 menit pada suhu 95 derajat. (12)
15. Dinginkan, dengan mengeluarkan slide dari decloaking dan disimpan pada suhu ruangan(14)
16. Setelah dingin , cuci dalam larutan PBS 2x masing-masing selama 5 menit(14)
17. Tandai slide dengan memberi lingkaran sekitar jaringan (14)
18. Atur slide pada baki slide (15)
19. Ambil satu per satu lalu tetesi Background Sniper lalu inkubasi selama 30 menit (16)
20. Buang larutan background sniper dengan cara ditiriskan pada tissu(16)
21. Tetesi Antibody Primer lalu inkubasi selama 1 jam pada suhu ruang(16)
22. Cuci PBS 2x selama 5 menit (17)
23. Tetesi Trekkie Universal lalu diamkan selama 30 menit(18)
24. Tiriskan pada tissue lalu tetesi Trekkkavidin-HRP lalu diamkan selama 30 menit
25. Cuci PBS 2x dengan cara merendam slide selama 5 menit
26. Sambil menunggu pencucian, buat larutan DAB dengan cara campurkan cromogen DAB 1 tetes + Substrat buffer 1 ml (dicampur dalam tabung bersih) lalu tetesi kejaringan
27. Amati jaringan jika sudah menunjukkan warna coklat, langsung
    dam ke dalam air selama 5 menit (20)
    lam dengan hematoxylin Meyer 5 menit (20)
    air mengalir selama 5 menit (21)
    drasi (Alkohol 70%, Alkohol 80%, Alkohol 96%) masing-masing 5
    t (22)

31. Clearing (Xylol I, Xylol II, Xylol III) (22)
32. Keringkan slide lalu tetesi dengan entelan lalu tutup dengan deck glass.(23)
33. Amati di Mikroskop (23)

9. TAHAPAN PROSEDUR IMMUNOHISTOKIMIA

		
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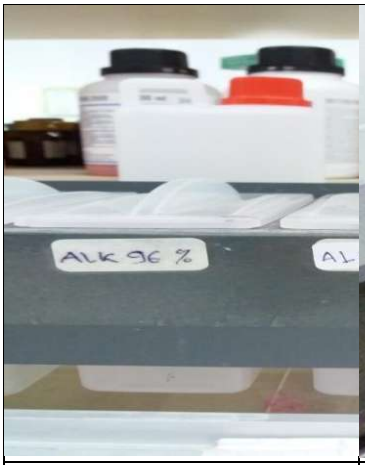		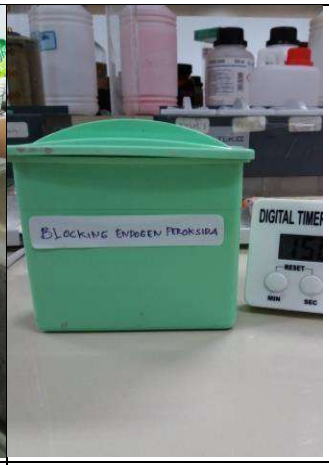
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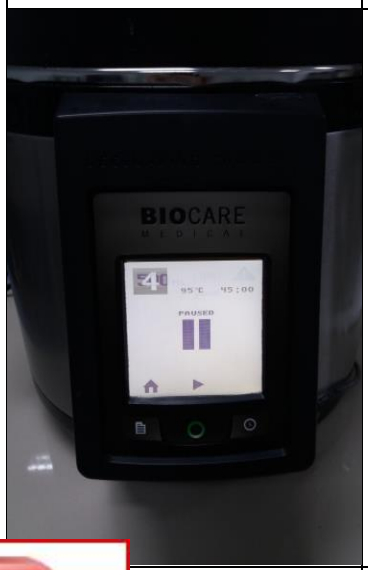		
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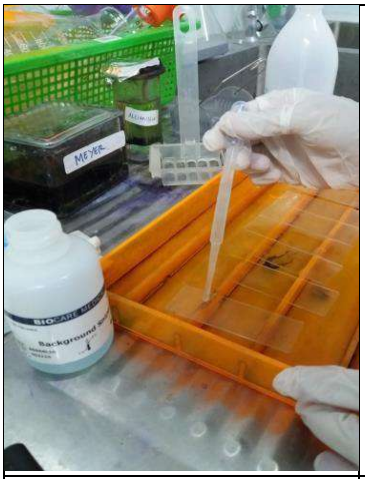

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10. Penjelasan Kerangka Konsep

Flavanoid yang merupakan salah satu komponen aktif kelor yang dikombinasikan dengan DDM yang memiliki kemampuan osteokonduksi dan osteoinduktif karena adanya growth factor berupa BMP yang dimasukkan kedalam soket bekas pencabutan sehingga menyebabkan aggregasi platelet yang melepaskan growth faktor dan sitokin. Sitokin memulai reaksi inflamasi yang didominasi oleh neutrofil yang kemudian digantikan oleh makrofag. Makrofag melepaskan growth faktor TGF yang salah satu anggotanya adalah BMP2. BMP 2 dapat menginduksi diferensiasi Mesencymal Stem Cell (MSC) menjadi osteoblas. Osteoblas menyebabkan terjadinya pembentukan tulang. Osteoblas juga menyebabkan ekspresi OPG meningkat. Kandungan bioaktif flavonoid yang menghambat NFkB menyebabkan RANKL menurun begitu pula mediator pro Inflammatory menurun. Ikatan RANKL yang menurun dan RANK menyebabkan NFkB menurun dan menyebab diferensiasi osteoklas menurun dan menyebabkan resorbsi tulang alveolar.