

## DAFTAR PUSTAKA

- Ahmed, A. *et al.* (2020) 'Pravastatin for early-onset pre-eclampsia: a randomised, blinded, placebo-controlled trial', *Bjog*, 127(4), pp. 478–488. doi:10.1111/1471-0528.16013.
- Araujo, J.A., Zhang, M. and Yin, F. (2012) 'Heme Oxygenase-1, Oxidation, Inflammation, and Atherosclerosis', *Frontiers in Pharmacology*, 3. doi:10.3389/fphar.2012.00119.
- Barber, A. *et al.* (2001) 'Heme oxygenase expression in human placenta and placental bed: reduced expression of placenta endothelial HO-2 in preeclampsia and fetal growth restriction', *FASEB journal: official publication of the Federation of American Societies for Experimental Biology*, 15(7), pp. 1158–1168. doi:10.1096/fj.00-0376com.
- Blackburn S (2015) *Maternal, Fetal, & Neonatal Physiology*.
- Brownfoot, F.C. *et al.* (2016) 'Effects of simvastatin, rosuvastatin and pravastatin on soluble fms-like tyrosine kinase 1 (sFlt-1) and soluble endoglin (sENG) secretion from human umbilical vein endothelial cells, primary trophoblast cells and placenta', *BMC pregnancy and childbirth*, 16, p. 117. doi:10.1186/s12884-016-0902-3.
- Conde-Agudelo, A. and Belizán, J.M. (2000) 'Risk factors for pre-eclampsia in a large cohort of Latin American and Caribbean women', *BJOG: an international journal of obstetrics and gynaecology*, 107(1), pp. 75–83. doi:10.1111/j.1471-0528.2000.tb11582.x.
- Costantine, M.M. *et al.* (2010a) 'Using pravastatin to improve the vascular reactivity in a mouse model of soluble fms-like tyrosine kinase-1-induced preeclampsia', *Obstetrics and Gynecology*, 116(1), pp. 114–120. doi:10.1097/AOG.0b013e3181e10ebd.
- Costantine, M.M. *et al.* (2010b) 'Using pravastatin to improve the vascular reactivity in a mouse model of soluble fms-like tyrosine kinase-1-induced preeclampsia', *Obstetrics and Gynecology*, 116(1), pp. 114–120. doi:10.1097/AOG.0b013e3181e10ebd.

- Costantine, M.M. *et al.* (2016) 'Safety and Pharmacokinetics of Pravastatin Used for the Prevention of Preeclampsia in High-Risk Pregnant Women: A Pilot Randomized Controlled Trial', *American journal of obstetrics and gynecology*, 214(6), p. 720.e1-720.e17. doi:10.1016/j.ajog.2015.12.038.
- Cudmore, M. *et al.* (2007) 'Negative regulation of soluble Flt-1 and soluble endoglin release by heme oxygenase-1', *Circulation*, 115(13), pp. 1789–1797. doi:10.1161/CIRCULATIONAHA.106.660134.
- Cudmore, M.J. *et al.* (2012) 'Resveratrol inhibits the release of soluble fms-like tyrosine kinase (sFlt-1) from human placenta', *American Journal of Obstetrics and Gynecology*, 206(3), p. 253.e10–15. doi:10.1016/j.ajog.2011.11.010.
- Duckitt, K. and Harrington, D. (2005) 'Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies', *BMJ : British Medical Journal*, 330(7491), p. 565. doi:10.1136/bmj.38380.674340.E0.
- Ferrier, K.E. *et al.* (2002) 'Intensive cholesterol reduction lowers blood pressure and large artery stiffness in isolated systolic hypertension', *Journal of the American College of Cardiology*, 39(6), pp. 1020–1025. doi:10.1016/S0735-1097(02)01717-5.
- Fox, K.A. *et al.* (2011) 'Effects of pravastatin on mediators of vascular function in a mouse model of soluble Fms-like tyrosine kinase-1-induced preeclampsia', *American Journal of Obstetrics and Gynecology*, 205(4), p. 366.e1–5. doi:10.1016/j.ajog.2011.06.083.
- Gajzlerska-Majewska, W., Bomba-Opon, D.A. and Wielgos, M. (2018) 'Is pravastatin a milestone in the prevention and treatment of preeclampsia?', *Journal of Perinatal Medicine*, 46(8), pp. 825–831. doi:10.1515/jpm-2017-0109.
- George, E.M. *et al.* (2011) 'Induction of heme oxygenase-1 attenuates sFlt-1-induced hypertension in pregnant rats', *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology*, 301(5), pp. R1495–R1500. doi:10.1152/ajpregu.00325.2011.
- George, E.M. and Granger, J.P. (2011) 'Mechanisms and potential therapies for preeclampsia', *Current Hypertension Reports*, 13(4), pp. 269–275. doi:10.1007/s11906-011-0204-0.

- George, E.M. and Granger, J.P. (2013) 'Heme oxygenase in pregnancy and preeclampsia', *Current opinion in nephrology and hypertension*, 22(2). doi:10.1097/MNH.0b013e32835d19f7.
- Granger, J.P. *et al.* (2001) 'Pathophysiology of Hypertension During Preeclampsia Linking Placental Ischemia With Endothelial Dysfunction', *Hypertension*, 38(3), pp. 718–722. doi:10.1161/01.HYP.38.3.718.
- Katsi, V. *et al.* (2017) 'The Role of Statins in Prevention of Preeclampsia: A Promise for the Future?', *Frontiers in Pharmacology*, 8, p. 247. doi:10.3389/fphar.2017.00247.
- Khashan, A.S. *et al.* (30 Jul 19) 'Preeclampsia and risk of end stage kidney disease: A Swedish nationwide cohort study', *PLOS Medicine*, 16(7), p. e1002875. doi:10.1371/journal.pmed.1002875.
- Kumasawa, K. *et al.* (2011) 'Pravastatin induces placental growth factor (PGF) and ameliorates preeclampsia in a mouse model', *Proceedings of the National Academy of Sciences of the United States of America*, 108(4), pp. 1451–1455. doi:10.1073/pnas.1011293108.
- Lee, T.-S. *et al.* (2004) 'Simvastatin induces heme oxygenase-1: a novel mechanism of vessel protection', *Circulation*, 110(10), pp. 1296–1302. doi:10.1161/01.CIR.0000140694.67251.9C.
- Marrs, C.C. and Costantine, M.M. (2017) 'Should we add pravastatin to aspirin for preeclampsia prevention in high- risk women?', *Clinical obstetrics and gynecology*, 60(1), pp. 161–168. doi:10.1097/GRF.0000000000000248.
- Meor Anuar Shuhaili, M.F.R. *et al.* (2017) 'Effects of Different Types of Statins on Lipid Profile: A Perspective on Asians', *International Journal of Endocrinology and Metabolism*, In Press(In Press). doi:10.5812/ijem.43319.
- Meyer, N. *et al.* (2021) 'Pravastatin Promotes Endothelial Colony-Forming Cell Function, Angiogenic Signaling and Protein Expression In Vitro', *Journal of Clinical Medicine*, 10(2). doi:10.3390/jcm10020183.
- Parikh, N.I. and Gonzalez, J. (2017) 'Preeclampsia and Hypertension', *JAMA internal medicine*, 177(7), pp. 917–918. doi:10.1001/jamainternmed.2017.1422.

- Ramma, W. and Ahmed, A. (2014) 'Therapeutic potential of statins and the induction of heme oxygenase-1 in preeclampsia', *Journal of Reproductive Immunology*, 101–102, pp. 153–160. doi:10.1016/j.jri.2013.12.120.
- Saad, A.F. *et al.* (2014) 'Effects of pravastatin on angiogenic and placental hypoxic imbalance in a mouse model of preeclampsia', *Reproductive Sciences (Thousand Oaks, Calif.)*, 21(1), pp. 138–145. doi:10.1177/1933719113492207.
- Tonelli *et al.* (2006) 'Effect of pravastatin on blood pressure in people with cardiovascular disease', *Journal of Human Hypertension*, 20(8), pp. 560–565. doi:10.1038/sj.jhh.1002036.
- Tsigos, C. *et al.* (2000) 'Stress, Endocrine Physiology and Pathophysiology', in Feingold, K.R. *et al.* (eds) *Endotext*. South Dartmouth (MA): MDText.com, Inc. Available at: <http://www.ncbi.nlm.nih.gov/books/NBK278995/> (Accessed: 13 July 2020).
- Tsur, A. *et al.* (2019) 'Pravastatin improves fetal survival in mice with a partial deficiency of heme oxygenase-1', *Placenta*, 75, pp. 1–8. doi:10.1016/j.placenta.2018.11.001.
- Weissgerber, T.L. and Mudd, L.M. (2015) 'Preeclampsia and Diabetes', *Current diabetes reports*, 15(3), p. 579. doi:10.1007/s11892-015-0579-4.
- Ye, Y.-C., Zhao, X.-L. and Zhang, S.-Y. (2015) 'Use of Atorvastatin in Lipid Disorders and Cardiovascular Disease in Chinese Patients', *Chinese Medical Journal*, 128(2), pp. 259–266. doi:10.4103/0366-6999.149226.
- Zenclussen, M.L. *et al.* (2015) 'Heme oxygenase-1 is critically involved in placentation, spiral artery remodeling, and blood pressure regulation during murine pregnancy', *Frontiers in Pharmacology*, 5, p. 291. doi:10.3389/fphar.2014.00291.

Lampiran 1. Penjelasan Penelitian Untuk Disetujui (Information for consent)

**FORM INFORMED CONSENT**

**Penjelasan Penelitian untuk Disetujui (*Information for consent*)**

Nama Peneliti : Andre Septian Putra

Judul Penelitian: PENGARUH PEMBERIAN PRAVASTATIN TERHADAP KADAR HEMEOKSIGENASE-1 PADA PASIEN RISIKO TINGGI PREEKLAMPSIA

- A. Tujuan penelitian & penggunaan hasilnya  
 Penelitian ini bertujuan untuk menilai efektifitas obat (Pravastatin) dalam menurunkan kadar hemeoksigenase-1 pada pasien risiko tinggi preeklamsia.
- B. Manfaat bagi peserta penelitian  
 Penemuan terapi baru untuk mencegah preeklamsia dengan menilai perubahan kadar hemeoksigenase-1 pada ibu yang memiliki risiko tinggi terhadap preeklamsia
- Metode dan prosedur kerja penelitian
- Penderita yang memenuhi kriteria inklusi dan eksklusi akan diberikan penjelasan mengenai penelitian ini, mulai tujuan penelitian, perlakuan, pengawasan, efek samping, risiko, pengambilan sampel dan lain-lain
  - Penderita akan ditawarkan untuk ikut berpartisipasi dalam penelitian ini, dan dijelaskan hak dan kewajiban sebagai partisipan penelitian
  - Jika penderita setuju menjadi partisipan penelitian ini, maka wajib menandatangani lembar persetujuan penelitian
  - Partisipan kemudian akan dilakukan randomisasi menjadi dua kelompok: yang menerima kombinasi pravastatin dan aspirin, serta yang menerima aspirin saja
  - Partisipan akan meminum obat sampai akhir kehamilan
  - Kadar hemeoksigenase-1 akan diperiksa sebelum dan setelah pemberian obat.
- C. Resiko yang mungkin timbul  
 Penelitian terkini menunjukkan bahwa pemberian pravastatin pada ibu hamil tidak meningkatkan risiko terjadinya kelainan bawaan pada janin.

1. Penelitian Data farmakologis Merck, dengan 477 ibu hamil yang mengkonsumsi statin. Tidak terbukti adanya peningkatan risiko kelainan bawaan pada janin pada ibu hamil yang mengkonsumsi simvastatin atau lovastatin (Kazmin A et al, 2007).
  2. Dari penelitian metaanalisis berskala besar, yang menilai efek pemberian statin pada ibu hamil trimester pertama. Tidak didapatkan peningkatan risiko kelainan bawaan janin, namun didapatkan sedikit peningkatan risiko keguguran pada trimester pertama (Zarek J, Koren G, 2014).
- D. Efek samping penelitian  
Konsumsi pravastatin dapat menimbulkan efek samping ringan (nyeri ulu hati, nyeri otot, nyeri dada, pusing, diare, nyeri kepala, batuk, bengkak, mual muntah, demam, kelelahan, sesak ringan, gejala flu), dan risiko efek samping berat (gangguan liver, dan kelainan otot). Dari penelitian Constatine (2016), tidak didapatkan peningkatan risiko terjadinya efek samping pada pemberian pravastatin.
- E. Tindak lanjut jika terjadi insiden saat dilaksanakan penelitian  
Apabila terjadi insiden akan dilakukan tindakan pengobatan sesuai standar yang biayanya ditanggung oleh peneliti.
- F. Jaminan kerahasiaan  
Identitas peserta penelitian, data, hasil penelitian dan semua yang berhubungan dengan penelitian ini akan dirahasiakan oleh tim peneliti.
- G. Hak untuk menolak menjadi subyek penelitian  
Subyek penelitian berhak menolak ikut serta dalam penelitian tanpa mempengaruhi perawatan selanjutnya.
- H. Partisipasi berdasarkan kesukarelaan dan hak untuk mengundurkan diri  
Subyek penelitian berpartisipasi secara sukarela, diberi kesempatan untuk menanyakan hal-hal yang belum jelas dan berhak mendapatkan jawaban yang memuaskan. Tiap saat dalam periode penelitian, subyek penelitian berhak mengundurkan diri dari penelitian.
- I. Subjek dapat dikeluarkan dari penelitian  
Bila subyek penelitian tidak mentaati instruksi yang diberikan oleh para peneliti, maka dapat dikeluarkan setiap saat dari penelitian ini.
- J. Penelitian ini dilakukan oleh

Makassar, .....

Yang memberi penjelasan

Yang menerima penjelasan

(.....)

(.....)

Saksi I

Saksi II

(.....)

(.....)

## Lampiran 2. Lembar Persetujuan Mengikuti Penelitian (Informed consent)

**FORM INFORMED CONSENT**  
**LEMBAR PERSETUJUAN MENGIKUTI PENELITIAN (*Informed consent*)**

Saya yang bertanda tangan dibawah ini :

Nama : .....

Umur : .....

Alamat : .....

Tlp / Email : .....

Sesudah mendengarkan penjelasan yang diberikan dan diberikan kesempatan untuk menanyakan yang belum dimengerti, dengan ini memberikan :

**PERSETUJUAN**

Mengikuti penelitian sebagai subyek penelitian dengan judul penelitian:  
**PENGARUH PEMBERIAN PRAVASTATIN TERHADAP KADAR HEMEOKSIGENASE-1 PADA PASIEN RISIKO TINGGI PREEKLAMPSIA**

dan sewaktu-waktu saya berhak mengundurkan diri.

Demikian persetujuan ini saya buat dengan penuh kesadaran dan tanpa paksaan.

Makassar,

.....

Yang memberi penjelasan

Yang membuat pernyataan

(.....)

(.....)

Saksi I

Saksi II

(.....)

(.....)



## Lampiran 3. Lembar Persetujuan Tindakan Medis

**LEMBAR PERSETUJUAN TINDAKAN MEDIS**

Saya yang bertanda tangan dibawah ini :

Nama :.....

Umur :.....

Alamat:.....

Tlp / Email :.....

Sesudah mendengarkan penjelasan yang diberikan dan diberikan kesempatan untuk menanyakan yang belum dimengerti, dengan ini memberikan :

**PERSETUJUAN**

Untuk dilakukan tindakan medis berupa:

.....  
.....

Dengan judul penelitian:

**PENGARUH PEMBERIAN PRAVASTATIN TERHADAP KADAR HEMEOKSIGENASE-1 PADA PASIEN RISIKO TINGGI PREEKLAMPSIA**

Subjek penelitian juga menyetujui bahwa sampel yang diambil akan dapat dilakukan pemeriksaan di laboratorium baik di dalam atau luar negeri.

Sewaktu-waktu saya berhak mengundurkan diri.

Demikian persetujuan ini saya buat dengan penuh kesadaran dan tanpa paksaan.

Makassar, .....

Yang Membuat Pernyataan

(.....)

Saksi 1

Saksi 2

(..... ) (.....)

## Lampiran 4. Lembar Pengumpulan Data Dasar Penelitian

**LEMBAR PENGUMPULAN DATA DASAR PESERTA PENELITIAN**

Nama : .....  
 Usia : .....  
 No Rekam Medis : .....  
 Rumah Sakit : .....  
 Alamat : .....  
 No Telpon : .....

**Kelompok** : Pravastatin - Kontrol

**Data Fisik, Antropologis, dan Etnografis**

Suku : Makassar-Bugis-Mandar-Toraja  
 Tempat kelahiran : .....  
 Tempat kelahiran orang tua : .....  
 Paritas : .....  
 Gravida : .....  
 Tinggi badan : ..... cm  
 Berat Badan : ..... kg  
 BMI : ..... kg/m<sup>2</sup>

**Riwayat Penyakit**

Hipertensi : + / -  
 Penyakit ginjal : + / -  
 Diabetes mellitus : + / - [jika +, Tipe 1 atau Tipe 2]  
 Penyakit vaskular kolagen (SLE, APS) : + / -  
 Riwayat Preeklamsia : + / -  
 Riwayat DM Gestasional : + / -

**Riwayat Obsetri**

(Berikan keterangan kehamilan keberapa dan usia kehamilan saat terjadinya)

Keguguran : + / - .....  
 Kematian janin dalam rahim : + / - .....  
 Keguguran yang diinduksi : + / - .....  
 Hipertensi Gestasional : + / - .....  
 Preeklamsia : + / - .....  
 Eklampsia : + / - .....  
 Sindroma HELLP : + / - .....

IUGR atau Bayi KMK : + / - .....  
 DM Gestasional memerlukan insulin : + / - .....  
 Persalinan preterm (< 37 minggu) : + / - .....  
 Kematian neonatal : + / - .....

### Riwayat Kehamilan Saat Ini

Tekanan darah saat pertama datang (*booking*) : ..... mmHg  
 Kehamilan multipel : + / -  
 Mola Hidatidosa : + / -  
 Plasenta hidrofik : + / -  
 Konsumsi obat anti hipertensi : + / -  
 Mendapat terapi SM : + / -  
 Mendapat terapi steroid untuk maturasi paru : + / -  
 Mendapat terapi obat anti tiroid : + / -

### Hasil pemeriksaan

Kadar profil lipid sebelum terapi :  
 Kadar profil lipid setelah terapi :  
 Kadar HO-1 darah sebelum terapi :  
 Kadar HO-1 darah setelah terapi :  
 RI doppler arteri uterina :  
 Fetal scan pada uk 20 - 24 minggu :  
 Doppler arteri umbilikal :  
 Biometri janin pada usia kehamilan 28 - 32 minggu :

### Diagnosis Preeklamsia

Data tekanan darah tertinggi selama kehamilan : .....  
 Data tekanan darah tertinggi saat persalinan : .....  
 Data tekanan darah tertinggi 48 jam setelah persalinan : .....  
 Proteinuria : + .....  
 Keterlibatan multiorgan (trombositopenia, peningkatan SGOT/SGPT, BUN-SK, kejang, persalinan preterm, IUGR, kematian fetal atau neonatal): ya - tidak .....

### Luaran Ibu

Mengalami Preeklamsia : + / -  
 Mengalami Preeklamsia Berat : + / -  
 Mengalami Hipertensi Gestasional : + / -  
 Kematian Ibu : + / - .....  
 Persalinan preterm < 37 minggu : + / -  
 Persalinan preterm < 34 minggu : + / -

Komplikasi Ibu : + / - [jika ya, sebutkan: edema paru, eklampsia, gagal ginjal akut, sindroma HELLP, tekanan darah  $\geq$  180/110 mmHg, DIC, CVA]  
 Lama Perawatan di RS : + / -  
 Metode persalinan : Per vaginam - Pervaginam dengan alat – SC

### **Luaran Janin**

Kematian fetal-neonatal : + / -  
 Morbiditas neonatal gabungan (IUFD, RDS, ICH, NEC, neonatal sepsis, IUGR) : + / -

Usia Kehamilan saat dilahirkan : .....  
 Berat Badan Bayi : ..... g  
 Panjang Badan Bayi : ..... cm  
 Ballard Score (jika ada) : ..... minggu  
 Lutzchenko Score (jika ada) : P .....  
 Kelainan kongenital : + / - .....  
 Tingkat perawatan : Bayi sehat - *intermediate care* - NICU  
 Lama perawatan di NICU : ..... hari  
 Pemakaian ventilator : + / -  
 Lama perawatan di RS : ..... hari  
 Mengalami gangguan dalam rahim (oligohidramnion berat, AEDV atau REDV pada arteri umbilikalis, abnormal NST) : + / -

### **Efek Samping Obat**

Nyeri kepala : + / -  
 Insomnia : + / -  
*Flushing skin* : + / -  
 Nyeri otot, atau kelemahan: + / -  
 Mengantuk berlebihan : + / -  
 Pusing : + / -  
 Mual muntah : + / -  
 Nyeri abdomen hebat : + / -  
 Kembung : + / -  
 Diare : + / -  
 Konstipasi : + / -  
 Bercak kulit : + / -

### **Efek Samping Berat Obat**

Miositis (keradangan otot) : + / -  
 Rhabdomyolisis : + / -

**Catatan:** Pemilihan jenis data dasar yang dikumpulkan berdasarkan pada standarisasi penelitian Preeklampsia yang dikeluarkan COLAB (*Global Pregnancy CoLaboratory*): *Strategy for Standardization of Preeclampsia Research Study Design* (Myatt L et al, 2014) dan luaran penelitian ini.

## Lampiran 5. Protokol Penelitian


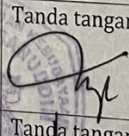

### PROTOKOL PENELITIAN

1. Tim peneliti melakukan skrining pada semua ibu hamil usia kehamilan 12 minggu – 19 minggu 6 hari di poliklinik
2. Ibu hamil yang memenuhi kriteria inklusi dan eksklusi akan ditawarkan untuk terlibat dalam penelitian ini
3. Tim peneliti memberikan penjelasan (*information for consent*) secara detail mengenai tujuan penelitian, prosedur penelitian, perlakuan, monitoring dan follow up pasien, hak dan kewajiban partisipan penelitian.
4. Jika pasien bersedia mengikuti penelitian ini, maka harus menandatangani lembar persetujuan penelitian (*informed consent*)
5. Kemudian tim peneliti akan mengambil data dasar partisipan dari wawancara dan pemeriksaan fisik (sesuai lampiran 4)
6. Tim peneliti kemudian akan membagi partisipan kedalam kelompok perlakuan atau kontrol berdasarkan randomisasi yang telah ditetapkan sebelumnya
7. Pembagian randomisasi kelompok dapat dilakukan sendiri oleh tim peneliti atau pihak farmasi rumah sakit
8. Setelah partisipan dimasukkan dalam salah satu kelompok, maka ia harus meminum obat tersebut selama kehamilan
9. Tim peneliti akan *memfollow up* dan memonitor partisipan selama kehamilan sesuai dengan jadwal pemeriksaan kehamilan standar
10. Adanya efek samping obat patut dievaluasi oleh tim peneliti, dan dicatat di lembar pengumpulan data (lampiran 4)
11. Partisipan akan diikuti sampai kelahiran dan pasca persalinan
12. Tim peneliti akan mencatat luaran maternal dan fetal-neonatal setelah partisipan melahirkan sebagai luaran penelitian ini
13. Hasil luaran penelitian dan data dasar partisipan akan dicatat dan dikompilasi di lembar kompilasi hasil penelitian (dalam bentuk file microsoft excel)



Nama Tim Peneliti (No Telp):

1. Dr. dr. Deviana Soraya Riu, SpOG (K) (0811460330)
2. dr. Rudy B. Leonardy, SpOG (K) (08124182638)
3. dr. Andre Septian Putra (082266609863)

## Lampiran 6. Rekomendasi Persetujuan Etik

 KEMENTERIAN PENDIDIKAN DAN KEBUDAYAAN UNIVERSITAS HASANUDDIN FAKULTAS KEDOKTERAN KOMITE ETIK PENELITIAN KESEHATAN RSPTN UNIVERSITAS HASANUDDIN RSUP Dr. WAHIDIN SUDIROHUSODO MAKASSAR Sekretariat : Lantai 2 Gedung Laboratorium Terpadu JL.PERINTIS KEMERDEKAAN KAMPUS TAMALANREA KM.10 MAKASSAR 90245. Contact Person: dr. Agussalim Bukhari.,MMed,PhD, SpGK TELP. 081241850858, 0411 5780103, Fax : 0411-581431			
<b>REKOMENDASI PERSETUJUAN ETIK</b>			
Nomor : 1143/UN4.6.4.5.31/ PP36/ 2019			
Tanggal: 29 Nopember 2019			
Dengan ini Menyatakan bahwa Protokol dan Dokumen yang Berhubungan Dengan Protokol berikut ini telah mendapatkan Persetujuan Etik :			
No Protokol	UH19080615	No Sponsor Protokol	
Peneliti Utama	<b>dr. Andre Septian Putra</b>	Sponsor	
Judul Peneliti	Pengaruh Pemberian Pravastatin Terhadap Kadar Hemeoksigenase I Pada Pasien Risiko Tinggi Preeklamsia		
No Versi Protokol	2	Tanggal Versi	<b>19 Nopember 2019</b>
No Versi PSP	2	Tanggal Versi	<b>19 Nopember 2019</b>
Tempat Penelitian	<b>Puskesmas Jumpandang Baru Makassar</b>		
Jenis Review	<input type="checkbox"/> Exempted <input type="checkbox"/> Expedited <input checked="" type="checkbox"/> Fullboard Tanggal 18 September 2019	Masa Berlaku <b>29 Nopember 2019</b> sampai <b>29 Nopember 2020</b>	Frekuensi review lanjutan
Ketua Komisi Etik Penelitian Kesehatan FKUH	Nama <b>Prof.Dr.dr. Suryani As'ad, M.Sc.,Sp.GK (K)</b>	Tanda tangan	
Sekretaris Komisi Etik Penelitian Kesehatan FKUH	Nama <b>dr. Agussalim Bukhari, M.Med.,Ph.D.,Sp.GK (K)</b>	Tanda tangan	
Kewajiban Peneliti Utama: <ul style="list-style-type: none"> <li>• Menyerahkan Amandemen Protokol untuk persetujuan sebelum di implementasikan</li> <li>• Menyerahkan Laporan SAE ke Komisi Etik dalam 24 Jam dan dilengkapi dalam 7 hari dan Laporan SUSAR dalam 72 Jam setelah Peneliti Utama menerima laporan</li> <li>• Menyerahkan Laporan Kemajuan (progress report) setiap 6 bulan untuk penelitian resiko rendah</li> <li>• Menyerahkan laporan akhir setelah Penelitian berakhir</li> <li>• Melaporkan penyimpangan dari protokol yang disetujui (protocol deviation / violation)</li> <li>• Mematuhi semua peraturan yang ditentukan</li> </ul>			

## Lampiran 7. Surat Izin Penelitian

 <b>RUMAH SAKIT UNHAS</b>	<b>SURAT IZIN PENELITIAN</b>	
	<b>Nomor:</b> 2757/UN4.24.1.2/PT.01.04/2021	<b>Tanggal</b> 08 Maret 2021
<b>FORMULIR</b> <b>2</b>  <b>BIDANG</b> <b>PENELITIAN DAN</b> <b>INOVASI</b>	Kepada Yth <b>Kepala Ruang Laboratorium Penelitian</b>	
<p>Dengan hormat,</p> <p>Dengan ini menerangkan bahwa peneliti/ mahasiswa berikut ini:</p> <p>Nama : dr. Andre Septian Putra</p> <p>NIM / NIP : C055171003</p> <p>Institusi : PPDS Ilmu Obstetri dan Ginekologi, Fakultas Kedokteran, Universitas Hasanuddin Makassar</p> <p>Kode penelitian : 210308_2</p> <p>Akan melakukan pengambilan data/ analisa bahan hayati:</p> <p>Terhitung : 08 Maret 2021 s/d 08 Juni 2021</p> <p>Jumlah Subjek/Sample : 70</p> <p>Jenis Data : Data Primer: Elisa</p> <p>Untuk penelitian dengan judul:</p> <p><b>"PENGARUH PEMBERIAN PRAVASTATIN TERHADAP KADAR HEMEOKSIGENASE-1 PADA PASIEN RISIKO TINGGI PREEKLAMPSIA"</b></p> <p>Harap dilakukan pembimbingan dan pendampingan seperlunya.</p> <p>Kepala Bidang Penelitian dan Inovasi</p> <p style="text-align: center;">   <b>Dr. Muhsin Firdaus Kasim, M.Sc</b>  <b>NIP. 198412012018073001</b> </p> <p><i>Catatan: Lembaran ini diarsipkan oleh Bidang Penelitian dan Inovasi</i></p>		

## Lampiran 8.

5th Edition, revised in May, 2020

**Elabscience®**

**(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSTICS !)**

### **Human HO1(Heme Oxygenase 1) ELISA Kit**

Synonyms: HO-1, HMOX1, HMOX1D, HSP32, Bk286b10

Catalog No : E-EL-H2172

96T

This manual must be read attentively and completely before using this product.

If you have any problems, please contact our Technical Service Center for help (info in the header of each page).

Phone: 240-252-7368(USA) 240-252-7376(USA)

Email: [techsupport@elabscience.com](mailto:techsupport@elabscience.com)

Website: [www.elabscience.com](http://www.elabscience.com)

Please refer to specific expiry date from label on the side of box.

Please kindly provide us with the lot number (on the outside of the box) of the kit for more efficient service.

5th Edition, revised in May, 2020

### **Intended use**

This ELISA kit applies to the in vitro quantitative determination of Human HO1 concentrations in serum, plasma and other biological fluids.

### **Specification**

●Sensitivity: 0.19 ng/mL

●Detection Range: 0.31-20 ng/mL

●Specificity: This kit recognizes Human HO1 in samples. No Significant cross-reactivity or interference between Human HO1 and analogues was observed.

●Repeatability: Coefficient of variation is < 10%.

### **Test principle**

This ELISA kit uses the Sandwich-ELISA principle. The micro ELISA plate provided in this kit has been pre-coated with an antibody specific to Human HO1. Standards or samples are added to the micro ELISA plate wells and combined with the specific antibody. Then a biotinylated detection antibody specific for Human HO1 and Avidin-Horseradish Peroxidase (HRP) conjugate are added successively to each micro plate well and incubated. Free components are washed away. The substrate solution is added to each well. Only those wells that contain Human HO1, biotinylated detection antibody and Avidin-HRP conjugate will appear blue in color. The enzyme-substrate reaction is terminated by the addition of stop solution and the color turns yellow. The optical density (OD) is measured spectrophotometrically at a wavelength of 450 nm ± 2 nm. The OD value is proportional to the concentration of Human HO1. You can calculate the concentration of Human HO1 in the samples by comparing the OD of the samples to the standard curve.



5th Edition, revised in May, 2020

**Kit components & Storage**

An unopened kit can be stored at 2-8°C for 1 month. If the kit is not used within 1 month, store the items separately according to the following conditions once the kit is received.

Item	Specifications	Storage
Micro ELISA Plate (Dismountable)	8 wells ×12 strips	-20°C, 6 months
Reference Standard	2 vials	
Concentrated Biotinylated Detection Ab (100×)	1 vial, 120 µL	
Concentrated HRP Conjugate (100×)	1 vial, 120 µL	-20°C(shading light), 6 months
Reference Standard & Sample Diluent	1 vial, 20 mL	4°C, 6 months
Biotinylated Detection Ab Diluent	1 vial, 14 mL	
HRP Conjugate Diluent	1 vial, 14 mL	
Concentrated Wash Buffer (25×)	1 vial, 30 mL	
Substrate Reagent	1 vial, 10 mL	4°C(shading light)
Stop Solution	1 vial, 10 mL	4°C
Plate Sealer	5 pieces	
Product Description	1 copy	
Certificate of Analysis	1 copy	

Note: All reagent bottle caps must be tightened to prevent evaporation and microbial pollution.  
The volume of reagents in partial shipments is a little more than the volume marked on the label, please use accurate measuring equipment instead of directly pouring into the vial(s).

**Other supplies required**

Microplate reader with 450 nm wavelength filter  
High-precision transfer pipette, EP tubes and disposable pipette tips  
Incubator capable of maintaining 37°C  
Deionized or distilled water  
Absorbent paper  
Loading slot for Wash Buffer

**Note**

1. Please wear lab coats, eye protection and latex gloves for protection. Please perform the experiment following the national security protocols of biological laboratories, especially when detecting blood samples or other bodily fluids.
2. A freshly opened ELISA Plate may appear to have a water-like substance, which is normal and will not have any impact on the experimental results.
3. Do not reuse the reconstituted standard, biotinylated detection Ab working solution, concentrated HRP conjugate working solution. The unspent undiluted concentrated biotinylated detection Ab (100×) and other stock solutions should be stored according to the storage conditions in the above table.
4. The microplate reader should have a 450(±10 nm) filter installed and a detector that can detect the wavelength. The optical density should be within 0–3.5.
5. Do not mix or use components from other lots.
6. Change pipette tips in between adding standards, in between sample additions, and in between reagent additions. Also, use separate reservoirs for each reagent.

**Sample collection**

**Serum:** Allow samples to clot for 2 hours at room temperature or overnight at 2–8°C before centrifugation for 15 min at 1000×g at 2–8°C. Collect the supernatant to carry out the assay. Blood collection tubes should be disposable and be non-endotoxin.

**Plasma:** Collect plasma using EDTA or heparin as an anticoagulant. Centrifuge samples for 15 min at 1000×g at 2–8°C within 30 min of collection. Collect the supernatant to carry out the assay. Hemolysed samples are not suitable for ELISA assay!

**Cell lysates:** For adherent cells, gently wash the cells with moderate amount of pre-cooled PBS and dissociate the cells using trypsin. Collect the cell suspension into a centrifuge tube and centrifuge for 5 min at 1000×g. Discard the medium and wash the cells 3 times with pre-cooled PBS. For each  $1 \times 10^6$  cells, add 150–250  $\mu\text{L}$  of pre-cooled PBS to keep the cells suspended. Repeat the freeze-thaw process several times until the cells are fully lysed. Centrifuge for 10 min at 1500×g at 2–8°C. Remove the cell fragments, collect the supernatant to carry out the assay. Avoid repeated freeze-thaw cycles.

**Tissue homogenates:** It is recommended to get detailed references from the literature before analyzing different tissue types. For general information, hemolysed blood may affect the results, so the tissues should be minced into small pieces and rinsed in ice-cold PBS (0.01M, pH=7.4) to remove excess blood thoroughly. Tissue pieces should be weighed and then homogenized in PBS (tissue weight (g): PBS (mL) volume=1:9) with a glass homogenizer on ice. To further break down the cells, you can sonicate the suspension with an ultrasonic cell disrupter or subject it to freeze-thaw cycles. The homogenates are then centrifuged for 5 min at 5000×g to get the supernatant.

**Cell culture supernatant or other biological fluids:** Centrifuge samples for 20 min at 1000×g at 2–8°C. Collect the supernatant to carry out the assay.

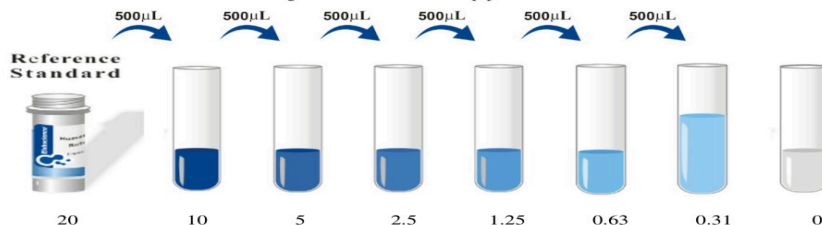
**Note for sample:**

1. Samples should be assayed within 7 days when stored at 2–8°C, otherwise samples must be divided up and stored at –20°C ( $\leq 1$  month) or –80°C ( $\leq 3$  months). Avoid repeated freeze-thaw cycles.
2. Please predict the concentration before assaying. If the sample concentration is not within the range of the standard curve, users must determine the optimal sample dilutions for their particular experiments.
3. If the sample type is not included in the manual, a preliminary experiment is suggested to verify the validity.
4. If a lysis buffer is used to prepare tissue homogenates or cell culture supernatant, there is a possibility of causing a deviation due to the introduced chemical substance.
5. Some recombinant protein may not be detected due to a mismatching with the coated antibody or detection antibody.

5th Edition, revised in May, 2020

**Reagent preparation**

1. Bring all reagents to room temperature (18–25°C) before use. Follow the Microplate reader manual for set-up and preheat it for 15 min before OD measurement.
2. **Wash Buffer:** Dilute 30 mL of Concentrated Wash Buffer with 720 mL of deionized or distilled water to prepare 750 mL of Wash Buffer. Note: if crystals have formed in the concentrate, warm it in a 40°C water bath and mix it gently until the crystals have completely dissolved.
3. **Standard working solution:** Centrifuge the standard at 10,000×g for 1 min. Add 1.0 mL of Reference Standard & Sample Diluent, let it stand for 10 min and invert it gently several times. After it dissolves fully, mix it thoroughly with a pipette. This reconstitution produces a working solution of 20 ng/mL. Then make serial dilutions as needed. The recommended dilution gradient is as follows: 20, 10, 5, 2.5, 1.25, 0.63, 0.31, 0 ng/mL. Dilution method: Take 7 EP tubes, add 500uL of Reference Standard & Sample Diluent to each tube. Pipette 500uL of the 20 ng/mL working solution to the first tube and mix up to produce a 10 ng/mL working solution. Pipette 500uL of the solution from the former tube into the latter one according to these steps. The illustration below is for reference. Note: the last tube is regarded as a blank. Don't pipette solution into it from the former tube.



4. **Biotinylated Detection Ab working solution:** Calculate the required amount before the experiment (100 µL/well). In preparation, slightly more than calculated should be prepared. Centrifuge the stock tube before use, dilute the 100× Concentrated Biotinylated Detection Ab to 1× working solution with Biotinylated Detection Ab Diluent.
5. **Concentrated HRP Conjugate working solution:** Calculate the required amount before the experiment (100 µL/well). In preparation, slightly more than calculated should be prepared. Dilute the 100× Concentrated HRP Conjugate to 1× working solution with Concentrated HRP Conjugate Diluent.

5th Edition, revised in May, 2020

**Assay procedure** (A brief assay procedure is on the 11<sup>th</sup> page)

1. Add the **Standard working solution** to the first two columns: Each concentration of the solution is added in duplicate, to one well each, side by side (100 µL for each well). Add the samples to the other wells (100 µL for each well). Cover the plate with the sealer provided in the kit. Incubate for 90 min at 37°C. Note: solutions should be added to the bottom of the micro ELISA plate well, avoid touching the inside wall and causing foaming as much as possible.
2. Remove the liquid out of each well, do not wash. Immediately add 100 µL of **Biotinylated Detection Ab working solution** to each well. Cover with the Plate sealer. Gently mix up. Incubate for 1 hour at 37°C.
3. Aspirate or decant the solution from each well, add 350 µL of **wash buffer** to each well. Soak for 1–2 min and aspirate or decant the solution from each well and pat it dry against clean absorbent paper. Repeat this wash step 3 times. Note: a microplate washer can be used in this step and other wash steps.
4. Add 100 µL of **HRP Conjugate working solution** to each well. Cover with the Plate sealer. Incubate for 30 min at 37°C.
5. Aspirate or decant the solution from each well, repeat the wash process for five times as conducted in step 3.
6. Add 90 µL of **Substrate Reagent** to each well. Cover with a new plate sealer. Incubate for about 15 min at 37°C. Protect the plate from light. Note: the reaction time can be shortened or extended according to the actual color change, but not more than 30min.
7. Add 50 µL of **Stop Solution** to each well. Note: Adding the stop solution should be done in the same order as the substrate solution.
8. Determine the optical density (OD value) of each well at once with a micro-plate reader set to 450 nm.

**Calculation of results**

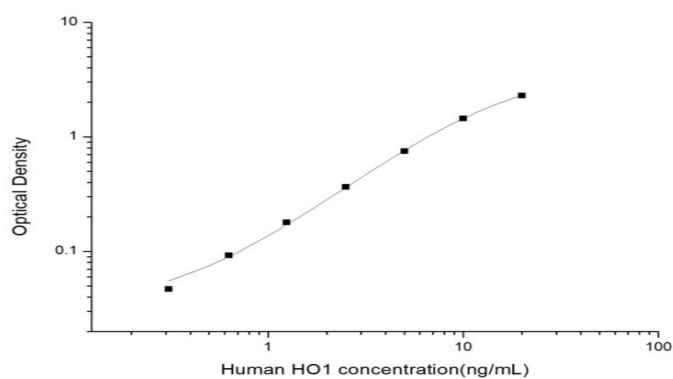
Average the duplicate readings for each standard and samples, then subtract the average zero standard optical density. Plot a four-parameter logistic curve on log-log graph paper, with standard concentration on the x-axis and OD values on the y-axis.

If the samples have been diluted, the concentration calculated from the standard curve must be multiplied by the dilution factor. If the OD of the sample surpasses the upper limit of the standard curve, you should re-test it with an appropriate dilution. The actual concentration is the calculated concentration multiplied by the dilution factor.

**Typical data**

As the OD values of the standard curve may vary according to the conditions of the actual assay performance (e.g. operator, pipetting technique, washing technique or temperature effects), the operator should establish a standard curve for each test. Typical standard curve and data is provided below for reference only.

Concentration(ng/mL)	20	10	5	2.5	1.25	0.63	0.31	0
OD	2.369	1.525	0.824	0.442	0.256	0.169	0.124	0.077
Corrected OD	2.292	1.448	0.747	0.365	0.179	0.092	0.047	-



5th Edition, revised in May, 2020

**Precision**

**Intra-assay Precision** (Precision within an assay): 3 samples with low, mid range and high level Human HO1 were tested 20 times on one plate, respectively.

**Inter-assay Precision** (Precision between assays): 3 samples with low, mid range and high level Human HO1 were tested on 3 different plates, 20 replicates in each plate.

Sample	Intra-assay Precision			Inter-assay Precision		
	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean(ng/mL)	1.09	3.15	9.57	0.99	3.28	9.54
Standard deviation	0.07	0.18	0.39	0.05	0.15	0.48
C V (%)	6.42	5.71	4.08	5.05	4.57	5.03

**Recovery**

The recovery of Human HO1 spiked at three different levels in samples throughout the range of the assay was evaluated in various matrices.

Sample Type	Range (%)	Average Recovery (%)
Serum (n=5)	94-107	101
EDTA plasma (n=5)	92-110	100
Cell culture media (n=5)	84-97	91

**Linearity**

Samples were spiked with high concentrations of Human HO1 and diluted with Reference Standard & Sample Diluent to produce samples with values within the range of the assay.

		Serum (n=5)	EDTA plasma(n=5)	Cell culture media(n=5)
1:2	Range (%)	89-103	99-111	88-103
	Average (%)	95	105	94
1:4	Range (%)	94-106	80-91	89-100
	Average (%)	100	86	95
1:8	Range (%)	88-100	85-101	86-97
	Average (%)	94	92	92
1:16	Range (%)	90-106	86-97	80-94
	Average (%)	97	92	87

5th Edition, revised in May, 2020

**Troubleshooting**

<b>Problem</b>	<b>Causes</b>	<b>Solutions</b>
Poor standard curve	Inaccurate pipetting	Check pipettes.
	Improper standard dilution	Ensure briefly spin the vial of standard and dissolve the powder thoroughly by gentle mixing.
	Wells are not completely aspirated	Completely aspirate wells in between steps.
Low signal	Insufficient incubation time	Ensure sufficient incubation time.
	Incorrect assay temperature	Use recommended incubation temperature. Bring substrate to room temperature before use.
	Inadequate reagent volumes	Check pipettes and ensure correct preparation.
	Improper dilution	
HRP conjugate inactive or TMB failure	Mix HRP conjugate and TMB, rapid coloring.	
Deep color but low value	Plate reader setting is not optimal	Verify the wavelength and filter setting on the Microplate reader.
		Open the Microplate Reader ahead to pre-heat.
Large CV	Inaccurate pipetting	Check pipettes.
High background	Concentration of target protein is too high	Use recommended dilution factor.
	Plate is insufficiently washed	Review the manual for proper wash. If using a plate washer, check that all ports are unobstructed.
	Contaminated wash buffer	Prepare fresh wash buffer.
Low sensitivity	Improper storage of the ELISA kit	All the reagents should be stored according to the instructions.
	Stop solution is not added	Stop solution should be added to each well before measurement.

**SUMMARY**

1. Add 100  $\mu$ L standard or sample to each well. Incubate for 90 min at 37°C.
2. Remove the liquid. Add 100  $\mu$ L Biotinylated Detection Ab. Incubate for 1 hour at 37°C.
3. Aspirate and wash 3 times.
4. Add 100  $\mu$ L HRP Conjugate. Incubate for 30 min at 37°C.
5. Aspirate and wash 5 times.
6. Add 90  $\mu$ L Substrate Reagent. Incubate for 15 min at 37°C.
7. Add 50  $\mu$ L Stop Solution. Read at 450 nm immediately.
8. Calculation of results.

**Declaration**

1. Limited by current conditions and scientific technology, we can't conduct comprehensive identification and analysis on all the raw material provided. So there might be some qualitative and technical risks for users using the kit.
2. The final experimental results will be closely related to the validity of products, operational skills of the operators and the experimental environments. Please make sure that sufficient samples are available.
3. To get the best results, please only use the reagents supplied by the manufacturer and strictly comply with the instructions!
4. Incorrect results may occur because of incorrect operations during the reagents preparation and loading, as well as incorrect parameter settings of the Micro-plate reader. Please read the instructions carefully and adjust the instrument prior to the experiment.
5. Even the same operator might get different results in two separate experiments. In order to get reproducible results, the operation of every step in the assay should be controlled.
6. Every kit has strictly passed QC test. However, results from end users might be inconsistent with our data due to some variables such as transportation conditions, different lab equipments, and so on. Intra-assay variance among kits from different batches might arise from the above reasons, too.











Lampiran 10. Kartu Kontrol Obat

# KARTU KONTROL OBAT

NAMA:  
 UMUR:  
 HPHT:  
 USIA KEHAMILAN:  
 TAFSIRAN PERSALINAN  
 KELOMPOK:

# 2018 2019

January							February							March						
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa
	1	2	3	4	5	6				1	2	3				1	2	3		
7	8	9	10	11	12	13	4	5	6	7	8	9	10	4	5	6	7	8	9	10
14	15	16	17	18	19	20	11	12	13	14	15	16	17	11	12	13	14	15	16	17
21	22	23	24	25	26	27	18	19	20	21	22	23	24	18	19	20	21	22	23	24
28	29	30	31				25	26	27	28				25	26	27	28	29	30	31

January							February							March						
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa
	1	2	3	4	5	6				1	2	3				1	2	3		
7	8	9	10	11	12	13	4	5	6	7	8	9	10	4	5	6	7	8	9	10
14	15	16	17	18	19	20	11	12	13	14	15	16	17	11	12	13	14	15	16	17
21	22	23	24	25	26	27	18	19	20	21	22	23	24	18	19	20	21	22	23	24
28	29	30	31				25	26	27	28				25	26	27	28	29	30	31

April							May							June								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
8	9	10	11	12	13	14	6	7	8	9	10	11	12	3	4	5	6	7	8	9		
15	16	17	18	19	20	21	13	14	15	16	17	18	19	10	11	12	13	14	15	16		
22	23	24	25	26	27	28	20	21	22	23	24	25	26	17	18	19	20	21	22	23		
29	30						27	28	29	30	31			24	25	26	27	28	29	30		

April							May							June								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
7	8	9	10	11	12	13	7	8	9	10	11	12	13	5	6	7	8	9	10	11		
14	15	16	17	18	19	20	14	15	16	17	18	19	20	12	13	14	15	16	17	18		
21	22	23	24	25	26	27	21	22	23	24	25	26	27	19	20	21	22	23	24	25		
28	29	30					28	29	30					26	27	28	29	30	31			

July							August							September								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
8	9	10	11	12	13	14	12	13	14	15	16	17	18	2	3	4	5	6	7	8		
15	16	17	18	19	20	21	19	20	21	22	23	24	25	9	10	11	12	13	14	15		
22	23	24	25	26	27	28	26	27	28	29	30	31		16	17	18	19	20	21	22		
29	30	31												23	24	25	26	27	28	29		

July							August							September								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
7	8	9	10	11	12	13	7	8	9	10	11	12	13	4	5	6	7	8	9	10		
14	15	16	17	18	19	20	14	15	16	17	18	19	20	11	12	13	14	15	16	17		
21	22	23	24	25	26	27	21	22	23	24	25	26	27	18	19	20	21	22	23	24		
28	29	30	31				28	29	30	31				25	26	27	28	29	30	31		

October							November							December								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
7	8	9	10	11	12	13	4	5	6	7	8	9	10	6	7	8	9	10	11	12		
14	15	16	17	18	19	20	11	12	13	14	15	16	17	13	14	15	16	17	18	19		
21	22	23	24	25	26	27	18	19	20	21	22	23	24	20	21	22	23	24	25	26		
28	29	30	31				25	26	27	28	29	30		27	28	29	30	31				

October							November							December								
Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa	Su	Mo	Tu	We	Th	Fr	Sa		
	1	2	3	4	5	6				1	2	3	4	5				1	2	3	4	5
7	8	9	10	11	12	13	4	5	6	7	8	9	10	6	7	8	9	10	11	12		
14	15	16	17	18	19	20	11	12	13	14	15	16	17	13	14	15	16	17	18	19		
21	22	23	24	25	26	27	18	19	20	21	22	23	24	20	21	22	23	24	25	26		
28	29	30	31				25	26	27	28	29	30		27	28	29	30	31				