

V. SIMPULAN DAN SARAN

A. Simpulan

Berdasarkan penelitian *Molecular Sexing* pada Accipitridae Menggunakan Metode Amplifikasi *Loop-Mediated Isothermal Amplification* (LAMP) dan *Polymerase Chain Reaction* (PCR) diperoleh kesimpulan sebagai berikut:

1. Metode PCR dapat mengamplifikasi gen *CHD-W* dan gen *CHD-Z* secara keseluruhan sebanyak 13 individu dari 7 spesies suku Accipitridae yang digunakan dengan keberhasilan identifikasi sebesar 100%. Metode LAMP dapat mengamplifikasi sampel betina maupun sampel jantan sehingga tidak dapat membedakan jenis kelamin betina atau jantan.
2. Primer LAMP ACCIW, ACCIZ, EBW, EB-Z dan NEO-W tidak dapat membedakan jenis kelamin betina atau jantan.

B. Saran

1. Perlu dilakukan optimasi lebih lanjut agar mendapatkan kondisi yang sesuai untuk reaksi amplifikasi LAMP
2. Disarankan untuk mendesain primer dengan teliti dan sesuai dengan protokol mendesain primer karena primer LAMP sangat berperan dalam keberhasilan amplifikasi.

DAFTAR PUSTAKA

- Archawaranon, M. 2004. Rapid Sexing Hill Mynah *Gracula religiosa* by Sex Chromosomes. *Biotechnology*, 3(2): 160–164.
- Astirin, O.P. 2000. Problems of Biodiversity Management in Indonesia. *Biodiversitas, Journal of Biological Diversity*, 1(1): 36–40.
- Basuki, O.P., Wijaya, K.D., Hidayat, A., Haryati, Y. dan Rakhman, Z. 2007. Laporan Teknis Pelepasliaran dan Monitoring Paska Pelepasliaran Elang Brontok (*Spizaetus cirrhatus*) di Taman Wisata Alam Danau Buyan-Danau Tamblingan. *Laporan Teknis*, 2: 1–70.
- BirdLife International 2004. *Important Bird Areas in Asia Key Sites for Conservation*. BirdLife International, Cambridge.
- BirdLife International 2016a. *Accipiter trivirgatus*. *The IUCN Red List of Threatened Species 2016*: e.T22695462A93510676.
<http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T22695462A93510676.en>.
- BirdLife International 2016b. *Haliaeetus leucogaster*. *The IUCN Red List of Threatened Species 2016*: e.T22695097A93489471.
<http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T22695097A93489471.en>.
- BirdLife International 2016c. *Nisaetus cirrhatus*. *The IUCN Red List of Threatened Species 2016*: e.T22732090A95042127.
<http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T22732090A95042127.en>.
- BirdLife International 2016d. *Spilornis cheela*. *The IUCN Red List of Threatened Species 2016*: e.T22695293A95221642.
<http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T22695293A95221642.en>.
- BirdLife International 2017. *Nisaetus bartelsi*. *The IUCN Red List of Threatened Species 2017*: e.T22696165A110050373.
<http://dx.doi.org/10.2305/IUCN.UK.2017-1.RLTS.T22696165A110050373.en>.
- Bramwell, R.K. 2003. Sexing Chicks in the Backyard Flock. *Avian Advice*, 5: 4–5.
- Brown, L.H. dan Amadon 1986. *Eagles, Hawks and Falcons of the World*. McGraw-Hill, New York.
- Centeno-Cuadros, A., Abbasi, I. dan Nathan, R. 2017. Sex Determination in the Wild: A Field Application of Loop-Mediated Isothermal Amplification Successfully Determines Sex Across Three Raptor Species. *Molecular Ecology Resources*, 17(2): 153–160.
- Cerit, H. dan Avanus, K. 2007a. Sex Determination by CHDW and CHDZ Genes of Avian Sex Chromosomes in *Nymphicus hollandicus*. *Turkish Journal of Veterinary and Animal Sciences*, 31(6): 371–374.

- Cerit, H. dan Avanus, K. 2007b. Sex Identification in Avian Species Using DNA Typing Methods. *World's Poultry Science Journal*, 63(01): 91–100.
- Chan, K.W., Liu, P.C., Yang, W.C., Kuo, J., Chang, C.L.T. dan Wang, C.Y. 2012. A Novel Loop-Mediated Isothermal Amplification Approach for Sex Identification of Columbidae Birds. *Theriogenology*, 78(6): 1329–1338.
- Christidis, L. 1985. A Rapid Procedure for Obtaining Chromosome Preparations from Birds. *Auk*, 102: 892–893.
- Cuadros, A.C., Tella, J.L., Delibes, M., Edelaar, P. dan Carrete, M. 2018. Validation of Loop-Mediated Isothermal Amplification for Fast and Portable Sex Determination Across the Phylogeny of Birds. *Molecular Ecology Resources*, 18(2): 251–263.
- Direktorat Jendral Perlindungan Hutan dan Konservasi Alam 2009. *Burung-burung Taman Nasional Baluran*. Balai Taman Nasional Baluran, Jawa Timur.
- Dubiec, A. dan Zagalska-Neubauer, M. 2006. Molecular Techniques for Sex Identification in Birds. *Biological Letter*, 23(1): 240–241.
- Ellegreen, H. 1996. First Gene on The Avian W-Chromosome (CHD) Provides a Tag for Universal Sexing of Non-Ratite Birds. *Proc Biol Sci*, 263(1377): 1635–1641.
- Fridolfsson, A.-K. dan Ellegren, H. 1999. A Simple and Universal Method for Molecular Sexing of Non-Ratite Birds. *Journal of Avian Biology*, 30(1): 116.
- Griffths, R., Double, M. C., Orr, K. dan Dawson, R.J.G. 1998. DNA Test to Sex Most Birds. *Molecular Ecology*, 7: 1071–1075.
- Handoyo, D. dan Rudiretna, A. 2001. Prinsip umum dan pelaksanaan Polymerase Chain Reaction (PCR). *Unitas*, 9(1): 17–29.
- Helander, B., Hailer, F. dan Vilà, C. 2007. Morphological and Genetic Sex Identification of White-tailed Eagle *Haliaeetus albicilla* Nestlings. *Journal of Ornithology*, 148(4): 435–442.
- Hsieh, K., Mage, P.L., Csordas, A.T., Eisenstein, M. dan Tom Soh, H. 2014. Simultaneous Elimination of Carryover Contamination and Detection of DNA with Uracil-DNA-Glycosylase-Supplemented Loop-Mediated Isothermal Amplification (UDG-LAMP). *Chemical Communications*, 50(28): 3747–3749.
- Khaerunnisa, I., Sari, E., Ulfah, M., Jakaria, J. dan Sumantri, C. 2013. Avian Sex Determination Based on Chromo Helicase DNA-binding (CHD) Genes Using Polymerase Chain Reaction (PCR). *Media Peternakan*, 36(2): 85–90.
- Kim, S.W., Choi, J.S., Sharma, N., Ko, Y.G., Do, Y.J., Byun, M., Seong, H.H., Park, S.B. dan Jeong, D.K. 2015. A Novel Approach for Determination of Chicken Sexing at an Early Stage of Development by Using Loop-mediated Isothermal Amplification Method. *Turkish Journal of Veterinary and Animal*

Sciences, 39(5): 583–588.

- Krisdianti 2018. Efektivitas Empat Pasang Primer untuk Identifikasi Jenis Kelamin Secara Molekuler pada Passeriformes. *Skripsi*, Universitas Atma Jaya Yogyakarta.
- Lee, P.L.M. 2017. DNA Amplification in the Field: Move over PCR, Here Comes LAMP. *Molecular ecology resources*, 17(2): 138–141.
- Lin, W.L., Lin, S.M., Lin, J.W., Wang, Y. dan Tseng, H.Y. 2015. Breeding Performance of Crested Goshawk *Accipiter trivirgatus* in Urban and Rural Environments of Taiwan. *Bird Study*, 62(2): 177–184.
- MacKinnon, J. 1994. *Panduan Lapangan Pengenalan Burung-burung di Jawa, Bali dan Kalimantan*. Puslitbang Biologi LIPI, Bogor.
- Menteri Lingkungan Hidup dan Kehutanan Republik Indonesia 2018. Peraturan Menteri Lingkungan Hidup dan Kehutanan Republik Indonesia NOMOR P.20/MENLHK/SETJEN/KUM.1/6/2018 Tentang Jenis Tumbuhan dan Satwa yang Dilindungi. 1–24.
- Nagamine, K., Hase, T. dan Notomi, T. 2002. Accelerated Reaction by Loop-Mediated Isothermal Amplification Using Loop Primers. *Molecular and Cellular Probes*, 16(3): 223–229.
- Notomi, T., Okayama, H., Masubuchi, H., Yonekawa, T., Watanabe, K., Amino, N. dan Hase, T. 2000. Loop-Mediated Isothermal Amplification of DNA. *Nucleic acids research*, 28(12): E63.
- Parida, M., Sannarangaiah, S., Dash, P.K., Rao, P.V.L. dan Morita, K. 2015. Loop Mediated Isothermal Amplification (LAMP): A New Generation of Innovative Gene Amplification Technique; Perspectives in Clinical Diagnosis of Infectious Diseases. *Reviews in medical virology*, 25(1): 2–23.
- Prawiradilaga, D.M. 1999. *Elang Jawa Satwa Langka*. Biodiversity Conservation Project, Bogor.
- Prawiradilaga, D.M., Muratte, T., Muzakkir, A., Inoue, T., Kuswandono, Adam, A.S., Ekawati, D., Afianto, M.Y., Hapsoro, Ozawa, T. dan Noriaki, S. 2003. *Panduan Survei Lapangan dan Pemantauan Burung-burung Pemangsa*. Biodiversity Conservation Project-JICA. Japan Internasional Cooperation Agency.
- Ramono, H.E.P., Ulyani, Y.E.N.I.A.R.M., Yartinilia, S. dan Iguchi, H. 2015. Pemodelan Distribusi Kesesuaian Habitat Singgah Sikep Madu Asia (*Pernis ptilorhynchus*) di Pulau Rupa Berdasarkan Data Satellite- Tracking. 20(1): 61–68.
- Reddy, A., Prakash, V. dan Shivaji, S. 2007. A Rapid, Non-Invasive, PCR-based Method for Identification of Sex of the Endangered Old World Vultures (White-Backed and Long-Billed Vultures): Implications for Captive Breeding Programmes. *Research communications*, 92(5): 659–662.

- Retnaningtyas, R.W., Hermadhiyanti, W., Rahayu, D.A. dan Listyorini, D. 2015. The Identification of The White-bellied Sea Eagle (*Haliaeetus leucogaster*) Based on Morphological Characteristics. *KnE Life Sciences*, 2(1): 588.
- Sambrook, J., Fritsch, E.F. dan Maniatis, T. 1989. *Molecular Cloning Laboratory Manual 3rd Ed.* Cold Spring Harbor Laboratory Press, New York.
- Setiadi, A.P. 2000. *Status, Distribusi, Populasi, Ekologi dan Konservasi Elang Jawa (Spizaetus bartelsi) di Jawa Barat Bagian Selatan.* Yayasan Pribumi Alam Lestari, Bandung.
- Sitohang, L.R. 2017. Moleculer Sexing pada Elang Jawa (*Nisaetus bartelsi* Stresemann, 1924) dan Elang Brontok (*Nisaetus cirrhatus* Gmelin, 1788) Hasil Sitaan BKSDA di Yogyakarta. *Skripsi*, Universitas Atma Jaya Yogyakarta.
- Sitorus, N.D. dan Ernowo, J.B. 2017. Studi Habitat dan Perilaku Burung Pemangsa Famili Accipitridae di SPTN 1 Tegaldlimo Taman Nasional Alas Purwo, Jawa Timur. *Journal IPB*, 278–285.
- Sozer, R., Nijman, V. dan Setiawan 1999. *Panduan Identifikasi Elang Jawa (Nisaeaetus bartelsi).* Biodiversity Conservation Project, Bogor.
- Suleman, E., Mtshali, M.S. dan Lane, E. 2016. Investigation of False Positives Associated with Loop-Mediated Isothermal Amplification Assays for Detection of *Toxoplasma gondii* in Archived Tissue Samples of Captive Felids. *Journal of Veterinary Diagnostic Investigation*, 28(5): 536–542.
- Supriatna, A.A. 2012. Current Status of Diurnal Raptors in Indonesia and Its Conservation Challenges. *Ornis Mongolica*, (1): 67–73.
- Swengel, S.R. 1996. *Special Techniques, C: Sex Determination In: Cranes: Their Biology, Husbandry, and Conservation.* National Biological Service/International Crane Foundation, United States of America.
- Wang, D.G., Brewster, J.D., Paul, M. dan Tomasula, P.M. 2015. Two Methods for Increased Specificity and Sensitivity in Moop-Mediated Isothermal Amplification. *Molecules*, 20(4): 6048–6059.
- Widiana, A., Iqbal, R.M. dan Yuliawati, A. 2017. Estimasi Luasan dan Perkembangan Daerah Jelajah Elang Brontok (*Nisaetus cirrhatus*) Pasca Rehabilitasi di Pusat Konservasi Elang Kamojang Garut Jawa Barat. *Jurnal Istek*, 10(2): 1–18.
- Wilfinger, W.W., Mackey, K. dan Chomcynski, P. 1997. Effect of pH and Ionic Strength on the Spectrophotometric Assessment of Nucleic Acid Purity. *Biotechniques*, 22(3): 474–481.
- Yuda, P. 2014. Detection of Avian Malaria by Plasmodium Loop-Mediated Isothermal Amplification of Confiscated Hawk-eagles in the Yogyakarta, Indonesia. *Presented at International Raptor Conference, 8th ARRCN Symposium, Pune, India, 6–9 February 2014.*