

“Studi *in Vitro* Kombinasi Herbal (Jahe, Kunyit, dan Temulawak) dan Probiotik (*Lactobacillus acidophilus* dan *Lactobacillus brevis*) sebagai Alternatif *Growth Promoter*”

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INTISARI

Antibiotic growth promoters (AGP) dalam pakan ternak telah banyak digunakan untuk pemacu pertumbuhan dan meningkatkan produktivitas ternak. Di sisi lain, penggunaan antibiotik secara rutin pada pakan memiliki kontribusi yang tinggi terhadap terjadinya resistensi antibiotik pada ternak maupun manusia. Pemerintah dan *World Health Organization* telah melarang penggunaan AGP untuk mengurangi resistensi dan residu antibiotik. Pemanfaatan herbal dan probiotik banyak digunakan sebagai pakan fungsional pada unggas, namun penggunaan kombinasi keduanya belum banyak diteliti. Tujuan dari penelitian ini adalah untuk mengetahui peran kombinasi jahe merah, kunyit, dan temulawak serta *Lactobacillus acidophilus* dan *L. brevis* sebagai alternatif pengganti AGP. Identifikasi ulang terhadap *Salmonella enteritidis*, *L. acidophilus*, dan *L. brevis* dilakukan dengan melihat morfologi koloni, morfologi sel, pengecatan Gram, dan sifat bakteri secara biokimiawi. Peran ekstrak segar, ekstrak produk, ekstrak etanol (1.56 % – 100%), dan ekstrak aqua (1.56 % – 50%) herbal terhadap pertumbuhan *S. enteritidis* (10^6 CFU/ml) serta *L. acidophilus* dan *L. brevis* (10^9 CFU/ml) dilakukan dengan metode difusi *disc*. Ekstrak herbal yang mampu menghambat patogen dan mendukung pertumbuhan kandidat probiotik dilakukan uji lanjut dengan mengukur densitas optikal (DO) media pertumbuhan menggunakan *microplate reader*. Kemampuan hambat kandidat probiotik terhadap patogen juga diuji menggunakan difusi *disc*. Uji adhesi kandidat probiotik dilakukan dengan mencampurkan kombinasi herbal terbaik dari uji sebelumnya dengan sel epitelial ileum-sekum ayam broiler (10^5 sel/ml), kemudian diinkubasi pada suhu ruang selama 60 menit, dilakukan pengecatan Gram, dan dihitung jumlah bakteri yang melekat tiap 50 sel epitel. Tiap uji dilakukan pengulangan sebanyak 3 kali. Hasil uji menunjukkan bahwa ekstrak etanol temulawak memiliki aktivitas daya hambat tertinggi terhadap *S. enteritidis*, diikuti oleh ekstrak etanol jahe merah, dan ekstrak aqua temulawak. Ketiga ekstrak tersebut pada konsentrasi 3.13% juga mampu mendukung pertumbuhan *L. acidophilus* dan *L. brevis* secara signifikan. Uji lanjut menunjukkan bahwa kombinasi ekstrak etanol jahe merah 3.13% dan ekstrak aqua temulawak 3.13% memiliki daya hambat terhadap *S. enteritidis* dan mampu mendukung pertumbuhan *L. acidophilus* (DO 0.18 ± 0.00) serta *L. brevis* (DO 0.21 ± 0.01) signifikan lebih baik daripada ekstrak individu, kontrol positif, maupun kontrol glukosa. *Lactobacillus acidophilus* dan *L. brevis* memiliki daya hambat yang rendah terhadap *S. enteritidis* (<6 mm), namun demikian kemampuan adhesi *L. acidophilus* (420.00 ± 28.21) dan *L. brevis* (259.33 ± 24.03) yang ditreatmen dengan kombinasi ekstrak terbaik jauh lebih besar daripada *S. enteritidis* (202.00 ± 14.00).

Kata kunci : kombinasi herbal, probiotik, AGP, *feed additive*

**Study in Vitro Combination of Herbs (Ginger, Turmeric, and Wild Ginger)
and Probiotics (*Lactobacillus acidophilus* and *Lactobacillus brevis*)
as an Alternative Growth Promoter**

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ABSTRACT

Antibiotic growth promoters (AGP) in animal feed have been widely used to spur growth and increase livestock productivity. On the other hand, regular use of antibiotics in the diet has a high contribution to the occurrence of antibiotic resistance in livestock and humans. The government and the World Health Organization have banned the using of AGP to reduce antibiotic resistance and residue. Utilization of herbs and probiotics are widely used as functional feeds in poultry, but the using of combination of both, herbs and probiotics, has not been studied yet. The purpose of this research is to know the role of combination of red ginger, turmeric, wild ginger, *Lactobacillus acidophilus* and *L. brevis* to substitute AGP. Re-identification of *Salmonella enteritidis*, *L. acidophilus*, and *L. brevis* were done by looking at the colonies morphology, cell morphology, Gram staining, and their biochemical characteristics. The role of fresh extract, product extract, ethanol extract (1.56% - 100%), and water extract (1.56% - 50%) of herbs on growth of *S. enteritidis* (10^6 CFU/ml), *L. acidophilus* and *L. brevis* (10^9 CFU/ml) were performed by the disc diffusion method. Herbal extracts which able to inhibit pathogens and supporting probiotic candidate growth were tested further by measuring optical density of growth media using microplate reader. Inhibitory ability of probiotics candidate to pathogens was tested also using disc diffusion. Adhesion test of probiotics candidate was performed by mixing the best herbal combinations from the previous test with epithelial cells of ileum-cecum broilers (10^5 cells/ml), then it was incubated at room temperature for 60 minutes, Gram stained, and calculated the number of bacteria attached to as many as 50 cells epithelium. Each tests were repeated 3 times. The test results showed that 3.13% wild ginger ethanol extract has the highest inhibitory activity on *S. enteritidis*, followed by red ginger ethanol extract, and wild ginger water extract at the same concentration. The three extracts were also able to support the growth of *L. acidophilus* and *L. brevis*. Further test showed that the combination of 3.13% red ginger ethanol extract and 3.13% wild ginger water extract had the best inhibitory effect to *S. enteritidis*. Those combination were able to support the growth of *L. acidophilus* (OD $0.18 \pm .00$) and *L. brevis* (OD $0.21 \pm .01$) better than one extracts alone, positive controls, as well as glucose control. *Lactobacillus acidophilus* and *L. brevis* had low inhibitory effect on *S. enteritidis* growth (<6 mm), however adhesion ability of *L. acidophilus* (420.00 ± 28.21) and *L. brevis* (259.33 ± 24.03) treated with that extract combination was larger than *S. enteritidis* (202.00 ± 14.00).
Keywords: combination herbs, probiotic, AGP, feed additive