Short communications

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Bandicoot Rat (*Bandicota bengalensis*): A Novel Reservoir of Pathogenic Bacteria at Poultry Farms, Rawalpindi/Islamabad, Pakistan

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> Abstract.- The prevalence of pathogenic bacteria in fecal matter, urine and blood of bandicoot rat (Bandicota *bengalensis*) inhabiting poultry farms has been studied. For this purpose, fifteen bandicoot rats were captured with live traps from poultry farms of Rawalpindi/Islamabad, Pakistan and examined for presence of bacterial species. The fecal matter of bandicoot rat was found to be contaminated with Escherichia coli (7%), Klebsiella pp. (27%), Proteus spp. (15%) and Salmonella spp. (20%). The Proteus spp (20%) and Salmonella spp. (7%) were also isolated from urine of bandicoot rat. This preliminary report showed that bandicoot rat serves as reservoir of bacterial infection of Escherichia coli, Klebsiella spp., Proteus spp. and Salmonella spp. at poultry farms.

Key words: Bandicoot rat, poultry farms, pathogen reservoir.

Bandicoot rats (*Bandicota bengalensis*) are commonly found in agricultural fields but recently small patches of population of this species have been observed at poultry farms located at the suburbs of Rawalpindi/Islamabad. To best of our knowledge, no report of bandicoot rat presence at poultry farms in Pakistan has appeared to date. The possible reasons for bandicoot rat to adopt new habitat are easy availability of feedstuffs, water, shelter and location of poultry farms in its natural habitat. It is well documented that the rats are common commensal pest (Meerburg *et al.*, 2006)

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which can damage the food products, buildings, stored products, and also serve as potential vector of pathogenic diseases in human and animals. Rats may transmit the bacteria through feces, urine, and hair remnants (Padula *et al.*, 2000; Meerburg *et al.*, 2006). Rat population at poultry farms can be a major reservoir of pathogenic bacteria which can transmit bacteria in the environment, food and animals (Rose *et al.*, 2000). It is pertinent to mention that rodent control measures have decreased bacterial infection in the poultry houses (Henzler *et al.*, 1998) which show a direct association between rats and the poultry diseases.

Screening of rodents for pathogenic bacterial infection is essential to determine the risk of bacterial transmission in poultry birds and products. Therefore, present study was designed to explore the prevalence of pathogenic bacteria in fecal matter, urine and blood of bandicoot rat inhabiting the poultry farms.

Material and methods

Fifteen bandicoot rats were captured through live traps from poultry farms of Rawalpindi/ Islamabad, Pakistan and immediately transported to Poultry Research Institute, Rawalpindi. Rats were euthanized with chloroform and dissected for collection of fecal matter, urine and blood samples for the presence of bacterial pathogens. All samples were taken individually and aseptically after dissection to avoid contamination from external sources (Pocock *et al.*, 2001).

To prepare the culture media, 50g of MacConkey agar was added to 1 liter of distilled water and heated to dissolve the entire agar. The culture media was autoclaved, cooled down and poured to Petri plates. The Petri plates were allowed to solidify and refrigerated for further use. Enrichment media (Selenite broth) was prepared by the same method as given above and stored in sterilized conical flasks.

Samples (fecal matter, urine and blood) were inoculated into enrichment media and incubated at 37°C over night. The inoculated tubes were then checked for turbidity as an indicator of bacterial growth. To obtain the pure isolates, samples were allowed to grow on the MacConkey agar plates at 37°C for 12-24 hours. After incubation period colonies were selected on the basis of their morphological characteristics and again cultured. Isolated bacteria were identified by gram-staining and biochemical tests.

Results and discussion

The data on prevalence of bacteria viz; *Escherichia coli, Klebsiella* spp., *Proteus* spp. and *Salmonella* spp. in fecal matter, urine and blood of bandicoot rat residing at poultry farms is given in Table 1. The fecal matter of bandicoot rat was found to be contaminated with *Escherichia coli* (7%), *Klebsiella* pp. (27%), *Proteus* spp. (15%) and *Salmonella* spp. (20%). The *Proteus* spp. (20%) and *Salmonella* spp. (7%) were also isolated from urine of bandicoot rat. However, no bacteria were isolated from blood samples of the rats.

To best of our knowledge, this is the first report of bandicoot rat presence at poultry farms as a reservoir of pathogenic bacteria. It is well documented that rats may become source of pathogenic bacteria at poultry farms (Meerburg et al., 2006) and can contaminate poultry products (Arsenault et al., 2007; Humphrey, 2003). Presence of pathogenic bacteria in fecal matter and urine of bandicoot rat (Escherichia coli, Klebsiella spp., *Proteus* spp. and *Salmonella* spp.) can be dangerous as horizontal transmission of pathogenic bacteria in rats populations are common and fast (Welch et al., 1941). This suggested that higher population of bandicoot rat at poultry farms may increase the risk of the bacterial infection in poultry birds and products. Therefore, suitable management strategies are required to eliminate the bandicoot rat at poultry farms.

References

- Arsenault, J., Letellier, A., Quessy, S., Morin, J.P. and Boulianne, M., 2007. Canada. J. Food Protect. 70: 1350-1359.
- Hanzler, D.J. and Opitz, H.M. 1992. Avian Dis., 36: 625-631.
- Humphrey, T.J., 1989. J. appl. Bact., 66: 112-126.
- Meerburg, B.G., Jacobs-reitsma, W.F., Wagenaar, J.A. and Kijlstra, A., 2006. *Appl. environ. Microbiol.*, **72**, 960– 962.
- Padula, P.J., Colavecchia, S.B., Martinz, V.P., Gonzalez Della Valle, M.O., Edelstein, A., Miguel, S.D., Russi, J., Riquelme, J.M., Colucci, N., Almiron, M. and Rabinovich, R.D., 2000. J. clin. Microbiol., 38: 3029-

3035.

- Pocock, M.J.O., Searle, J.B., Betts, W.B. and White, P.C.L., 2001. J. appl. Microbiol., **90**: 755–760.
- Rose, N., Beaudeau, F., Drouin, P., Toux, J., Rose, V. and Colin, P., 2000. *Prev. Vet. Med.*, **39**: 9–20.
- Welch, H., Ostrolenk, M. and Bartram, M.T., 1941. Am. J. Publ. Hlth., **31**: 332-340.

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First Record of *Binodoxys basicurvus* Shujauddin, 1973 (Hymenoptera: Braconidae: Aphidiinae) From the Punjab Province of Pakistan

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Abstract. Aphid parasitoid, *Binodoxys basicurvus* Shujauddin, 1973, is recorded for the first time from the Punjab Province of Pakistan. Illustrated redescription of the species is provided with its host range and distribution in Pakistan.

Key words: *Binodoxys,* hymenoptera, aphidiinae.

Binodoxys is a genus of Aphidiinae parasitoids. It has two sets of tubercles of various sizes and shapes, on the basis of which it can be separated from the *Trioxys* genus (Kavallieratos *et al.*, 2001).Various species of this genus have been recorded from different parts of the world like Israel (Mackauer, 1959; 1960; Mecheloff and Rosen, 1993), Southeastern Europe (Kavallieratos *et al.*, 2001), Japan (Takada, 1968) and Czechoslovakia (Kavallieratos and Lykouressis, 1999; Kavallieratos

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et al., 2002a,b, 2004a,b,c, 2005, 2008a,b; Tomanović *et al.*, 2003, 2006, 2008). Starỳ (1979) reported 5 species from the Central Asian Area while Starỳ and Schlinger (1967) reviewed 8 species of *Binodoxys* from the Far Eastern part of the world.

In neighboring countries, various workers have reported this genus (Shujaudin, 1973; Takada and Rishi, 1980; Bhagat, 1982), including the record of 16 species by Raychaudhuri (1990) from India. So far 3 species of *Binodoxys* have been reported from Pakistan (Starỳ *et al.*, 1998) out of a total of 30 Aphidiinae parasitoid species (Starỳ *et al.*, 2006; Naeem *et al.*, 2005). Keeping in view this situation, various surveys were conducted during 2005-08 for the collection of aphid parasitoids. Many samples of *Binodoxys basicurvus* Shujauddin were collected. In this manuscript, this species, which is the first record from Pakistan, is being redescribed. Its distribution in Pakistan and host range have also been described.

Materials and methods

Samples of parasitized aphids together with plant parts and live aphids were collected from various public parks from 6 districts of the Punjab Province of Pakistan during 2005-2008 and then transferred to the laboratory in plastic bags. The materials were subsequently placed in plastic rearing boxes covered with cloth mesh for ventilation. The female aphids were killed, preserved in 70 percent alcohol for identification. Mummified aphids were placed in gelatinized capsules in order to observe the emergence hole and coloration of mummies.

The emerged wasps were collected using an aspirator and stored in 99% ethyl alcohol for future work. The parasitoids were identified according to reliable keys (Shujauddin, 1973; Raychaudhuri, 1990). The illustrations were prepared using a Nikon microscope (SMS-1500, with 30x 1-11.25x magnification). Measurements were taken using ocular micrometer in Noif microscope (XSZ 107BN. with 10X10X magnification). The morphological terminology used in this paper follows Sharkey and Wharton (1997).

Binodoxys basicurvus Shujauddin, 1973. Indian J.

Entoml., 35: 9-10.

Description of the female Head (Fig. 1A)

Smooth with sparsely distributed setae. Ocellar triangle acute, oval shaped. Head wider than mesosoma. Eyes large, oval shaped, sparsely setose and strongly convergent towards clypeus. Temple as wide as $\frac{1}{2}$ eye length, distinctly wider than $\frac{1}{2}$ eye width. Gena nearly as wide as $\frac{1}{4}$ th of longitudinal eye diameter. Clypeus with 4 long setae. Interocular line twice the transfacial line. Antennae 11 segmented, thickened toward apex. F1 somewhat longer than F2, almost three and a half times longer than wide; flagellar segments 3-8 as long as first flagellar segment, almost three times longer than wide.

Mesosoma

Mesoscutum smooth, with long sparsely distributed long hairs and distinct notaulices at the ascending part. Propodeum (Fig. 1D), 1.2-1.4 times as wide as long at spiracles, with distinct pentagonal areola, upper areola with 7-8 setae and lower with 2 setae. Stigma (Fig. 1B) broadly triangular, 1.61 times as long as metacarpus.

Metasoma

Pctiole (Fig. 1C) with both primary (spiracular) and secondary tubercles, about 2.7 times as long as wide at spiracles and with distinct central longitudinal carina. Genitalia as in (Fig. 1E). Prongs (Fig. 1F) slightly curved beyond basal third with 3 long dorsal and 2 short apical setae.

Coloration

Head dark brown. Ocelli yellow. Maxillary and labial palpi yellow. Scape, pedicle, F1, yellow. F4-9 brown. Prothorax, mesosterna and propodeum yellowish, remaining mesosoma brown. Wing venation brown. Abdomen brown in coloration except tergites 7, 8 and sternites 6, 7 which are yellow. Prongs yellow.

Male

Similar to female. Antennae 13 segmented. Body dark brown (excepting legs and petiole which are brownish).



Fig. 1. External morphology of *Binodoxys* basicurvus and the parasitized aphid. A, head anterior view showing scape, pedicle and F1-F2; B, anterior view of the Fore wing; C, Metasomal tergum I; D, propodeum; E, female genitalia; F, emergence hole of the adult parasitoid on posterio-dorsal part of the mummified aphid (*Aphis gossypii*).

Material examined

Aphis gossypii Glover (Hemiptera: Aphididae) on *Hibiscus rosa- sinensis*, Rawalpindi, 28.02.08, 4 \bigcirc and 2 \Diamond ; Islamabad, 04.03.05, 3 \bigcirc and 1 \Diamond ; Gujranwala , 16-3-06, 3 \bigcirc and 1 \Diamond ; Lahore, 21-3-07, 5 \bigcirc and 2 \Diamond ; Multan, 20-3-06, 2 \bigcirc and 1 \Diamond .

Aphis craccivora Koch (Hemiptera: Aphididae) on *Hibiscus rosa- sinensis*, Rawalpindi, 28.02.08, 2° and 2° ; Islamabad, 04.03.05, 2° ; Gujranwala , 16-3-06, 1° ; Faisalabad, 13-3-08, 4° and 2° ; Lahore, 21-3-07, 2° and 1° ; Multan, 20-3-06, 2° and 1° .

Remarks

Specimens collected from Pakistan were compared with description given by Shujaudin (1973) and Raychaudhuri (1990) and found to be morphologically similar excepting negligible color variation with reference to place of collection.

This species was firstly recoded from India by Shujauddin in 1973. Raychaudhuri (1990) mentioned its distribution from India from Uroleucon sonchi, Uroleucon sonchi (L.), Aphis gossypii and A. longisetosa Basu (Hemiptera: Aphididae). In Pakistan, it emerged from the mummies of Aphis gossypii and A. craccivora on Hibiscus rosa-sinensis L. (Malvaceae). Mummies (Fig. 1F) are light blackish brown to straw colored. Emergence hole usually at dorsum between the siphunculi.

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References

- Bhagat, R.C., 1982. Entomon, 7: 103-105.
- Kavallieratos, N. G. and Lykouressis, D.P., 1999. Boll. Lab. Ent. Agric. Filippo Silvestrii, 55: 93-104.
- Kavallieratos, N.G., Lykouressis, D.P., Sarlis, G.P., Stathas, G.J., Sanchis Segovia, A. and Athanassiou, C.G., 2001. *Phytoparasitica*, **29**: 306-340.
- Kavallieratos, N.G., Athanassiou, C.G., Stathas, G.J. and Tomanović, Ž., 2002a. *Phytoparasitica* **30**: 365-377.
- Kavallieratos, N. G., Stathas, G.J., Athanassiou, C.G. and Papadoulis, G.T.H., 2002b. *Phytoparasitica*, **30**: 231-242.
- Kavallieratos, N. G. and Lykouressis, D. P., 2004a. Isr. J. Ent.,

34: 75-82.

- Kavallieratos, N. G., Stathas, G. J. and Tomanović, Ž., 2004b. Biologia, 59: 191-196.
- Kavallieratos, N.G., Tomanović, Ž, Starý, P., Athanassiou, C.G., Sarlis, G. P., Petrović, O., Niketić, M. and Anagnou-Veroniki, M., 2004c. *Appl. Ent. Zool.*, **39**: 527-563.
- Kavallieratos, N.G., Tomanović, Ž., Athanassiou, C. G. Starý, P., Žikić, V., Sarlis, G. P. and Fasseas, C., 2005. Can. Ent., 137: 516-531.
- Kavallieratos, N. G., Tomanović, Ž., Starý, P and Emmanouel, N. E., 2008a. Flor. Entomol. 91: 179-191.
- Kavallieratos, N. G., Tomanović, Ž., Starý, P. and Mitrovski Bogdanović, A., 2008b. Zootaxa, **1793**: 47-64
- Mackauer, M., 1959. Beitr. Ent., 9: 866-873.
- Mackauer, M., 1960. Beitr. Ent., 10: 137-160.
- Mechelof, E. and Rosen, D., 1993. Trioxys Binodoxys. Isr. J. Ent., 27: 31-47.
- Naeem, M., Shehzad, F. and Khan, M.R., 2005. *Ent. Monogr. Maga.*, Humana Press, U. S. A. 141: 219-226.
- Raychaudhuri, D., 1990. Aphidiids (Hymenoptera) of Northeast India. Indra Publ. House, India, pp. 152.
- Sharkey, M. J. and Wharton, R.A., 1997. In: Manual of the New World genera of the family Braconidae (Hymenoptera) (eds. R.A. Wharton, R.A. Marsh and Sharkey, M.J.), pp. 19-37. International Society of Hymenopterists, Special Publication 1, Washington, pp. 439.

- Starý, P. and Schlinger, E.I., 1967. A revision of the Far East Asian Aphidiidae (Hymenoptera). Ser. Ent. 3. Dr. W. Junk. The Hague, pp. 204.
- Starý, P., 1979. Trans.Czechosl. Acad. Sci., Ser. math. nat. Sci., 89: 1-116
- Starý, P., Naumann–Etienne, K. and Remaudiere, G., 1998. *Parasitica.*, **54**: 3-21.
- Starý, P., 2006. Aphid parasitoids of the Czech Republic (Hymenoptera: Braconidae: Aphidiinae). Academia, Praha, pp. 432.
- Takada, H., 1968. Insecta Matsum., 30: 67-124.
- Takada, H. and Rishi, N. D., 1980. Kontyů Tokyo, 48: 234-240.
- Tomanović, Ž., Kavallieratos, N.G., Athanassiou, C.G. and Stanisavljević, L. Ž., 2003. Annl. Soc. Ent. France, 39: 343-353.
- Tomanovic, Ž., Kavallieratos, N. G., Starý, P., Petrovic, O., Tomanovic, S. and Jovanovic, S., 2006. J. Plant Dis.Protect., 113: 174-180.
- Tomanović, Ž., Starý, P., Kavallieratos, N. G., Petrović, A., Niketić, M. and Vučetić, A., 2008. *Zootaxa*, **1781**: 20-30.

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Research Econolyzer: A Software for Conducting Economic Analysis of Research Data

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> Traditionally, Abstract.pest management experiments in specific and agricultural experiments in general involve testing of various treatments or interventions to make recommendations that are based on the treatments/technologies that lead to a maximum increase in crop yield. Such recommendations, however, may not always be economically viable for farmers. Economic analysis of the research data can provide an estimate of risks and profitability of the proposed technologies before making recommendations for the farmers. Marginal analysis is one of such techniques which can assist research workers in decision making. A software has been developed to facilitate calculations involved and getting automated results for marginal rate of return (MRR). Along with the values of MRR, the software provides the values of residuals to assist in selecting the appropriate technology.

Key words: Economic analysis, marginal analysis, marginal rate of return.

Most pest management experiments in specific and agricultural experiments in general involve testing of various treatments or interventions in order to find one that leads to the increase in yield/quality of agricultural produce. Traditionally, recommendations are based on the treatments/ technologies that lead to a maximum increase in yield. However such recommendations may not be economically viable for farmers. Economic analysis of the research data can provide an estimate of risks and profitability of the proposed technologies before making recommendations for the farmers. Marginal analysis is one of such techniques which can assist research workers in decision making. The analysis calculates marginal rate of return (MRR) which is

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then compared with a minimum acceptable rate of return. The analysis justifies investment in higher cost technologies as long as they give a rate of return higher than the minimum acceptable rate of return as compared with low cost technologies (the word technologies is used interchangeably with treatments in this manuscript).

Perrin *et al.* (1988) has provided detailed guidelines on the steps involved in conducting marginal analysis. Based on the same guidelines, a software has been developed to facilitate calculations involved and getting automated results for MRR. Along with the values of MRR, the software provides the values of residuals to assist in selecting the appropriate technology. A maximum of eleven technologies or treatments in a field experiment, can be compared.

Input variables

Cost of inputs

The analysis does not take into consideration the total costs involved with the use of a certain technology. It rather considers the costs that vary within the technologies being tested. The inputs that are similar in all the technologies being tested are not considered. For example, if a researcher wants to compare five different chemical sprays for insect pest control under similar agronomic conditions, the costs of insecticide and its application will be considered. Other costs associated with land preparation, seed, fertilizer and irrigation (similar for all technologies) will not be used in the analysis.

Value of field output

Quantity and unit price of field output are entered to calculate farm income. If any marketing costs are involved like transportation to the market etc., these are to be subtracted from the market price before entering the data. Although major crop yield (like grain yield in case of cereals) is the main field output, sometimes by-products (*e.g.* straw) also contributes to the farm income and can be entered as field output as a result of applying a certain technology. If the produce is of variable quality, prices of different quality grades can be used for analysis.

Yield adjustment

As recommended by Perrin et al. (1988),

there is a provision of downward yield adjustment of 0-20%. It is done because yields from on-farm experiments are usually higher than those expected from farmers' fields.

Minimum acceptable rate of return

Minimum acceptable rate of return can be used an input for calculating residuals.

Output of analysis

The output produce by software includes partial budget, dominance analysis, marginal rate of return and residuals.

Partial budget

It involves calculation of net benefit calculated by subtracting the total costs that vary from the gross field benefit for each technology. The Gross benefit is calculated by multiplying the adjusted yield of the main produce or by-product by the market price.

Dominance analysis

Dominance analysis ensures that any experimental treatment that is more expensive must compensate its cost in the form of net benefit. In order to do this, the treatments are first sorted on ascending order on the basis of total costs that vary and then compared with their respective net benefits. If the net benefit of an experimental treatment is lesser or equal to that of another treatment which is lower in cost, it is said to be dominated and is excluded from further analysis.

Marginal rate of return

Marginal rate of return (MRR) is the ratio of marginal benefit to marginal cost for each experimental treatment. Marginal benefit is the change in net benefit when moving to the next costlier experimental treatment. Similarly marginal cost is the change in cost when moving to the next costlier experimental treatment. In order to do this, the treatments (excluding the dominated ones) are arranged in ascending order on the basis of their costs. Starting with the fist experimental treatment (the one with the lowest cost), the marginal benefit is computed by subtracting its benefit from the next higher experimental treatment (the second one in this case). Marginal cost is also calculated in a similar fashion by computing a difference of its cost from the next higher cost experimental treatment. Marginal rate of return is then calculated by dividing marginal benefit of each experimental treatment by its marginal cost which then expressed as percentage. MRR is not an absolute value for each experimental treatment. It rather is relative estimate which compares the change in benefit when switching from one technology to another one which is higher in cost.

The marginal rate of return indicates the gain, a producer can expect, when changing from one practice to another. For example if MRR for switching from technology A to B is 130%, it implies that for each dollar invested in the Technology B, the farmer can expect to get back one dollar plus an additional return of 1.3 dollars.

 Table I. An example of calculating marginal rate of return,

Net benefit % MRR	Total costs that vary	Pest control technology
450	0	А
800 350	100	В
996 178	210	С
1020 26	300	D
99617102026	210 300	C D

Before recommending a technology to the farmers, it is important to estimate the minimum MRR that would be sufficient for a farmer to adopt the new practice/technology. As suggested by Perrin *et al.* (1988), in majority of situations the minimum rate of return acceptable to the farmers will be 50 to 100%. If the technology is new to the farmer and requires learning new skill, a 100% minimum rate of return is a reasonable estimate. If a technology simply represents adjustment in current farming practice then a minimum rate of return as low as

50% may be acceptable. Table I is an example to illustrate this point.

In the above example moving from technology A (doing nothing) to technology B that cost Rs. 100 gives the maximum MRR (350%) but if the farmer continue to invest more and adopts technology C he can still get 178% return for his investment. Now let us consider moving to technology D. Although it has the maximum net benefit, a farmer will get only 26% more on the extra investment as compared with technology C. Since this is less than the acceptable level of minimum MRR, the technology should not be recommended to the farmers. Based on the above example, it is technology C that should be recommended to the farmers.

Residuals

Residuals can be used as criteria for selecting the appropriate treatment. The value indicates a difference between net benefit and cost of the investment. Residual for each treatment is calculated by subtracting the return that farmers require (minimum rate of return of return multiplied by the total costs that vary) from the net benefit (Perrin *et al.*, 1988). The treatment with the highest residuals is the one that should be recommended to the farmers. The software is available for free download at http://www.nifa.org.pk/Software.htm

Reference

Perrin, R., Anderson, J., Winkelmann, D. and Moscardi, E., 1988. From Agronomic Data to Farmer Recommendations: An Economic Training Manual. CIMMYT: Mexico, D.F. Available online at http://www.cimmyt.org/.

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