Azotobacter chroococcum were about 88.7% and 61.5%, respectively (table 3). The amount of nitrogen fixed per gram of carbon source was 11.5 mg by *Klebsiella* and 8.5 mg by *Enterobacter* as compared to 14.7 mg by *A. chroococcum*. Interestingly, the *Enterobacter* BH 25 showed higher nitrogenase activity than *Klebsiella* BH 124. However, the nitrogen fixed per gram of carbon source was more by the later bacterium. This was subsequently found to be due to the fact that the *Enterobacter* BH 25 possesses conventional hydrogenase and loses much energy in wasteful hydrogen production (data unpublished).

The counts of nitrogenase positive colonies, made in this study, are mainly of those bacteria which were growing aerobically on agar surface. The oxygen sensitive bacteria, however, were not studied in the present studies because of limitations in isolation and characterization of such bacteria. Also precise condition under which these bacteria express nitrogenase activity are not well known.

Our earlier studies with this crop showed that under dryland conditions, *Pseudomonas azotogenesis* was the main aerobic associative N_2 fixing bacteria[1], under irrigated conditions, however, there was a change in the associative forms and the major taxonomic types, found to be associated with bajra, were *klebsiella* sp. and *Enterobacter* sp.

12 October 1982

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GUT BACTERIAL FLORA OF COWPEA WEEVILS

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THE cowpea weevils*, *Callosobruchs analis* (Fabricius) and *C. maculatus* (Fabricius) (Coleoptera: Bruchidae) are serious pests of peas, beans and pulses[1,2]. Though bacteria associated with different groups of insects have been surveyed, the knowledge of bacteria associated with the storage insects[3,4] is scanty. In the present work it is aimed to report the bacteria associated with the digestive tract of two storage insect pests, *C. analis* and *C. maculatus*. This appears to be the first report of bacteria from cowpea weevils.

*Identified by Commonwealth Institute of Entomology, London.

The samples of pea and blackgram along with the infested cowpea weevils, *C. analis* and *C. maculatus* were collected from various godowns and maintained in the laboratory in clean sterilised transparent boxes. Living adult insects were randomly chosen and subjected to surface disinfection[5]. Effectiveness of the technique was decided by dipping some of the randomly chosen pretreated insects in nutrient broth which turned turbid within 24 hr with contaminated insects; and these were discarded.

After surface disinfection, hundred adults of each of the two insect pests were dissected aseptically and the guts obtained were grouped in 20 sets separately, for further study. Each set (having 5 guts) was homogenised in a known quantity of peptone broth and was incubated at 30 ± 1°C for 48-72 h, which served as the original stock suspension. Later, out of this suspension, 1 ml broth was serially diluted six times. One ml of such dilutent was pourplated, in duplicate, on each of the five media; nutrient agar, macConkey agar, sodium azide agar, blood agar, glucose agar, for the quantitative estimation of microflora, and was incubated at 30°C for 48-72 hr. Thus an average of 40 plate counts from each media was used for the calculation of the gut population. The bacterial populations were identified[6,7] and analysed statistically.

Results indicated that a total of eighteen bacterial types were found associated with the gut of the two cowpea weevils. These represented *Bacillaceae, Micrococcacea, Pseudomonadaceae, Enterobacteriaceae*, and *Flavobacteriaceae*. They were *B. cereus, B. circulans, B. subtilis, B. megaterium, B. coagulans, B. polymyxa, B. licheniformis, Micrococcus roseus, M. varians, M. luteus, Pseudomonas aeruginosa, Aerobacter aerogenes, A. cloacae, Citrobacter intermedius, C. freundii, Klebsiella pneumoniae, Proteus mirabilis* and *Flavobacterium lutescens*. Of the bacteria isolated, *A. aerogenes, A. cloacae, C. intermedius, C. freundii, Klebsiella pneumoniae* and *Ps. aeruginosa* are reported to be human pathogens[8,9]. *B. subtilis* is a food spoiling and food poisoning organism[7]. *A. aerogenes, A. cloacae, P. mirabilis, Ps. aeruginosa, B. subtilis, B. cereus* and *B. megaterium* are reported to be potential insect pathogens[10]. *M. roseus, M. varians, M. luteus, B. licheniformis, B. coagulans, B. circulans, B. polymyxa* and *Flavobacterium lutescens*

are common contaminats from soil, dust, water and various other sources thus might have accidental entry into the insect gut. Storage insects are known to transfer and spread viable bacterial spores to the stored peas and pulses they infest, by carrying them externally or through feces[11]. Further, the infections are also reported from human and animal consumers of such contaminated food[12,13]

The bacterial counts / ml / 5 guts revealed that gut bacterial population varied depending on the culture media. The counts from *C. analis* on sodium azide agar, macConkey agar, blood agar, glucose agar and nutrient agar was 88.0 ± 6.8., 167.0 ± 15.5., 224.5 ± 16.7., 459.3 ± 28.1., and 483.5 ± 37.5 respectively, while the same from *C. maculatus* was 24.4 ± 3.2., 151.5 ± 13.5., 141.8 ± 13.9., 411.3 ± 26.8 and 511.0 ± 27.6 respectively.

Thus, cowpea weevils, *C. analis* and *C. maculatus,* as they harbour several bacterial species including some that are human pathogens and a few that are food spoiling and food poisoning organisms, frequently can contaminate, damage and spoil stored peas and pulses and also can cause health problems in consumers.

The authors are thankful to Dr. M. Nagaraj, the Vice-Chancellor, Gulbarga University, Gulbarga, for his encouragement and for the facilities.

24 September 1982; Revised 8 November 1982

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TRYPANOSOMA RUPICOLI (SP. NOV.) FROM A HILLSTREAM FISH *NEMACHEILUS RUPICOLA* (HORA)

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TRYPANOSOMES of fish have been reported from all over the world[1,2]. From the Indian sub-continent itself 20 species of the trypanosomes have been described[3-8] But so far no report exists on the haematozoan parasites from the cold Himalayan realm of this country. The present paper reports the occurrence of trypanosomes from a freshwater, hillstream, teleost.

Two specimens of the host fish, *Nemacheilus rupicola* (Hora), harbouring the trypanosomes were obtained from the river Kosi, near Kosi station, 11km north of Almora. Blood was drawn from the caudal vein of the fish, at the spot, blood smears prepared, air dried, numbered and brought to the laboratory. Each slide was stained either with Leishman's or Geimsa's stain. Presence of trypanosomes was noted under the high power microscope and then camera lucida drawings made of the parasite, under oil immersion objective[7,8] and morphometric characters recorded.

Diagnosis and description: Body of the parasite appears stumpy or elongated (figures 1,2,3&5), typically curved in crescentic shape in most of the forms. Posterior end beak shaped and not sharply pointed, while the anterior end is sharply tapering Cytoplasmic contents of the cell body took light azurophilic stain, granulation scant and fine Vacuoles were present in almost all forms Myonemes not seen.