corresponding increase in solar radiation, the afternoon maximum becomes more prominent.

Evening and early night maxima are the result of convective activity. During the day, surface heating leads to increased convective activity which produces instability in the atmosphere and leads to katabatic winds during the night. Such katabatic winds from Garo, Khasi and Jayantiya Hills might produce rainfall in the evening and early night at Gauhati.

Table 1 shows that the average hourly rainfall per rainy day for the different months is a maximum in June and a minimum in September. The monthly rainfall normals for the monsoon period also show an almost similar behaviour. The coefficient of variation of average hourly rainfall is a maximum in September. The curves also indicate that only about 30% of the daily rainfall occurs in the late morning and afternoon hours (1000–2100 hr IST). Rainfall during the day hours (0600–1800 hr IST) is also observed to be less than that during the night hours.

Conclusion

Diurnal variation patterns of rainfall are different for each of the monsoon months at Gauhati. In June, August and September they show prominent peaks in the very early morning or late night which might be due to the effect of radiational cooling of the top of the cloud layers. During September, the higher amount of solar radiation received seems to play an important role in producing the afternoon maximum. The evening and early night maxima, specially in June and July, might be produced by katabatic winds from the neighbouring hilly areas.

The intensity of hourly rainfall is maximum in June and minimum in September. The present results may be considered to be tentative, as the data for only five years have been used in this study.

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SIMPLE DIAGNOSTIC TECHNIQUE FOR PLANT DISEASES OF MYCOPLASMAL ETIOLOGY

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MYCOPLASMAS which are also referred to as wall-less bacteria have been widely reported to parasitise on several plant hosts[1]. In affected plants the mycoplasma-like organisms (MLO)/mycoplasma-like bodies (MLB) occur as phloem delimited obligate parasites and induce diseases "yellows" of economic concern[2–5]. A major constraint in diagnosis of plant mycoplasma is that, except the spiroplasmas, they cannot be cultured in artificial media[1,6,7]. The association of MLO in host plants can be reliably detected by electron microscopy[1,5] and fluorescence microscopical technique[8] but they require elaborate procedures, expensive equipments and reagents. Therefore, a simple technique for quick diagnosis of mycoplasmal diseases is desirable. A diagnostic technique involving selective stains for plant diseases of viral etiology has been demonstrated[9]. In the detection of cultures of animal mycoplasmas and plant spiroplasmas on agar media the Dienes' stain is commonly used[6,10,11]. Plant and animal mycoplasmas

have great similarities[1]. This paper reports the utility of Dienes' stain for diagnosis of plant diseases of mycoplasmal etiology.

A stock solution of Dienes' stain was prepared by dissolving azure II (1.25 g), methylene blue (2.5 g), maltose (10 g) and sodium carbonate (0.25 g) in 100 ml distilled water[6,11] and filtered through Whatman No. 1 filter paper. A series of dilutions of the stain (0.05-0.50% v v) was also prepared. Representative samples of plants affected with MLO in India[4] *viz.*, spike of sandal (*Santalum album*), little leaf of periwinkle (*Vinca rosea*), little leaf of brinjal (*Solanum melongena*), phyllody of sesamum (*Sesamum indicum*) and yellow dwarf of rice (*Oryza sativa*) were collected for tests. Samples of the plant diseases of virus, bacterial and fungal etiology were also included in the study to test their reaction to the stain and their details are as follows:

Virus diseases: Tristeza of citrus (*Citrus* sp.), sterility mosaic of red gram (*Cajanus cajan*), mosaic of bean (*Dolichos lab lab*), vein-clearing of bhendi (*Hibiscus esculentus*), mosaic of chilli (*Capsicum annuum*) and bunchy top of banana (*Musa* sp.).

Bacterial diseases: Rice (*O. sativa*) bacterial blight (*Xanthomonas campestris* pv. *oryzae*), cotton (*Gossypium* sp.) bacterial blight (*X. campestris* pv. *malvacearum*), tomato (*Lycopersicon esculentum*) wilt (*Pseudomonas syringae* pv. *tomato*), citrus (*Citrus* sp.) canker (*X. campestris* pv. *citri*) and chilli (*C. annum*) leaf spot (*X. campestris* pv. *vesicatoria*).

Fungal diseases: Pearl millet (*Pennisetum typhoides*) downy mildew/green-ear (*Sclerospora graminicola*), grape vine (*Vitis vinifera*) downy mildew (*Plasmopara viticola*), sugarcane (*Saccharum officinarum*) red rot (*Colletotrichum falcatum*), rice (*O. sativa*) blast (*Piricularia oryzae*) and cocoa (*Theobroma cacao*) leaf spot (*Colletorichum gloeosporioides*).

In all instances, the healthy samples of the appropriate plants were used as control.

Stem and leaf parts of the samples were cut by free-hand sectioning using a single edge razor blade. The sections, suspended in distilled water, were transferred to the stain at different dilutions to react for 10-15 min. Subsequently, the stain was withdrawn by repeated washings and the sections resuspended in distilled water. The sections were mounted individually in distilled water on clean slides and covered with cover slip. The colour reactions of xylem, phloem and cortical regions of the sections were examined under a microscope at low and high magnifications.

In sections of all the samples the xylem was coloured turquoise blue and the cortex pale purplish blue. The phloem of plants affected by MLO stained dark/deep blue whereas none of the diseases due to viruses, bacteria or fungi exhibited phloem staining. All the mycoplasmal diseases showed a similar and positive reaction to Dienes' stain. Under higher magnifications, groups of dark bodies with blue coloured contents, distributed regularly in sieve elements and scattered in phloem arrays could be resolved in mycoplasmal diseases. In contrast, the phloem of healthy plants and those affected by viruses, bacteria or fungi remained unstained. Among the different dilutions of the stain tested, the dilution of 0.2% appeared to be optimum for the diagnosis of mycoplasmal diseases.

Dienes' stain has been reported to stain distinctly blue the colonies of animal mycoplasmas[10] and plant spiroplasmas[6,11] in agar media. The stain neither can act on viruses[2] nor retained by bacteria[6,11]. Dienes' stain distinguishes plant tissues affected by MLO from healthy ones by deeply staining the sieve elements. In plant hosts the MLO/MLB are restricted to phloem cells and the biology of plant and animal mycoplasmas are in great similarity[1]. The dark blue bodies observed in phloem of tissues affected by MLO appear due to uptake of Dienes' stain by phloem (sieve elements) protoplast possessing the parasitic mycoplasmal bodies[2,12]. The stain differentiated mycoplasmal infected tissues from healthy ones and gave negative reaction to the diseases of other etiology. It is interpreted that Dienes' stain can be used as a simple diagnostic tool for plant diseases of mycoplasmal etiology.

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RESIDUAL EFFECT OF SEVIN® ON THE ACETYL CHOLINESTERASE ACTIVITY OF THE NERVOUS SYSTEM OF EARTHWORM *PONTOSCOLEX CORETHRURUS*

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ABSTRACT

The effect of Sevin® on the AchE activity of the nervous system of earthworm *P. corethrurus* is concentration dependent. Concentrations below 100 ppm are tolerant to worms and increase the activity. Above 250 ppm level in soils, Sevin inhibit the activity. The AchE activity in the nervous preparations of earthworm and cockroach is inhibited with increasing concentrations of Sevin *in vitro*.

THE mortality of earthworm *P. corethrurus* to carbamates like Sevin® is dependent on the concentration of Sevin[1] although the worm tolerates the presence of Sevin in soils[2-4]. In the present study it is tested whether the carbamate like Sevin® inhibits the AchE (1-naphthyl-*n*-methyl carbamate) (EC 3.1.1.7) activity of the earthworm just as it does in insects[5,6].

Cement 'culture pots' (60 × 75 × 75 cm) were used for the study; each pot had 6 kg of fine sand, and 2 kg of clay-loam mixed with Sevin which ranged from 0 to 375 ppm. Earlier work indicated that Sevin above 375 ppm was not tolerant to worms[1]. Therefore, 20 different concentrations below this value were chosen. Into each pot twenty adult earthworms, *P. corethrurus* (with fully developed clitellum) were introduced. After one month they were recovered from the soil of the pot and grouped into five categories, according to the concentration of the Sevin mixed to the pot-soil. The worms recovered from pots without Sevin (0 ppm) served as control. The other four groups were from the pots containing (a) 37.5–50 ppm, (b) 75–100 ppm, (c) 150–200 ppm and (d) 250–375 ppm of Sevin.

The ventral nerve cord of the worm was excised after dissecting the worm in ice-cold physiological saline (0.6% NaCl). Homogenates of the tissue (1%) was prepared in 0.1 M phosphate buffer (pH 7.4) centrifuged at 600 *g* and the supernatant was used for the enzyme assay. The protein of the homogenate was estimated according to Lowry *et al.*[7] The AchE (EC 3.1.1.7) activity of the homogenate was estimated colorimetrically[9] and expressed as millimoles Ach hydrolized/mg protein/min.

The effects of Sevin *in vitro* on the AchE activities of cerebral ganglion extracts of cockroach as well as earthworm were tested in the same manner. The assay mixture consisted of 1 ml of the homogenate and 1 ml of Sevin in 0.1 M PO_4 buffer pH 7.4.

Exposure to Sevin upto 100 ppm in soil is congenial in accelerating the nerve cord AchE activity in the earthworm.

Above this concentration, Sevin showed significant inhibitory effects on AchE activity (figure 1), higher the concentration, more is the inhibitory effect. The

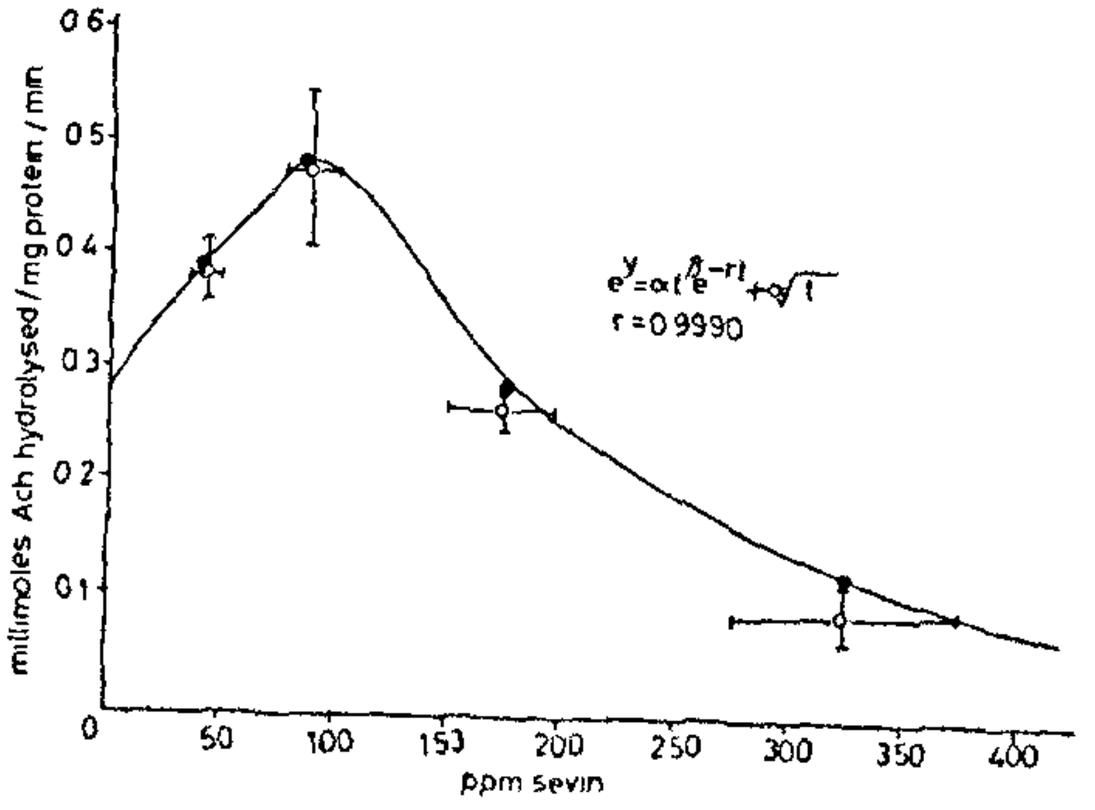

Figure 1. AchE activity in nervous system of earthworm *P. corethrurus* exposed to different concentrations of Sevin®. Plots on y-axis are mean ± S.D. of 10 observations. Variations of Sevin in 20 soil samples are indicated on x-axis.