derivatives are calculated by data of Lyman[5] and the equations of Culberson and Pytkowicz[6]. K'_B is apparent dissociation constant of boric acid.

It has been shown earlier[1] that total CO_2, TCO_2, corrected to salinity, total alkalinity and preformed CO_2(Co) is denoted by TCO_2'' and has linear relation with normalized AOU as

$$TCO_2'' = Co + 0.384\ (AOU). \qquad (2)$$

Considering (1) and (2), one can compute the corrected pH (25°) which is expected to show non-linear dependence with AOU. However, the corrected pH (25°) has linear dependence with normalized AOU. The normalised AOU is defined as

$$AOU_{normalized} = \frac{AOU \times 35}{salinity},$$

A new term called "$AOU_{relative}$" can be defined as

$$AOU_{relative} = \frac{(TCO_2'')_{pHi} - (TCO_2'')_{8.2}}{0.384}, \qquad (3)$$

where $(TCO_2'')_{pHi}$ is the value of TCO_2'' corresponding to a particular pHi. TCO_2'' was assigned the lowest value at zero $AOU_{relative}$ and at pH 8.2. $(TCO_2'')_{8.2}$ is total corrected CO_2 at pH 8.2. The theoretical pH curve can be calculated by

$$TCO_2'' = Co + 0.384\ AOU$$

$$= \left[TA_n - \frac{T_B - K'_B}{a_H + K'_B}\right]\left[\frac{a_H^2 + a_H K'_1 + K'_1 K'_2}{a_H K'_1 + 2K'_1 K'_2}\right]. \qquad (4)$$

In (4), TA_n is the normalized value of total alkalinity. In Central Arabian sea, the normalized value of TA_n (total alkalinity normalized to salinity and depth) is 2.771 meq kg^{-1} of sea water as reported elsewhere[2].

$AOU_{relative}$ obtained from (3) was plotted against pH in figure 1. The theoretical curve calculated from (4) was then superimposed over the experimental points in the figure. The points overlapping in the figure are removed for clarity. The theoretical curve was shifted along the x-axis until the best fit was obtained. A computer program developed on Apple II was employed for the purpose.

The standard deviation σ between the observed and the calculated pH values (269 points) was 0.015 unit.

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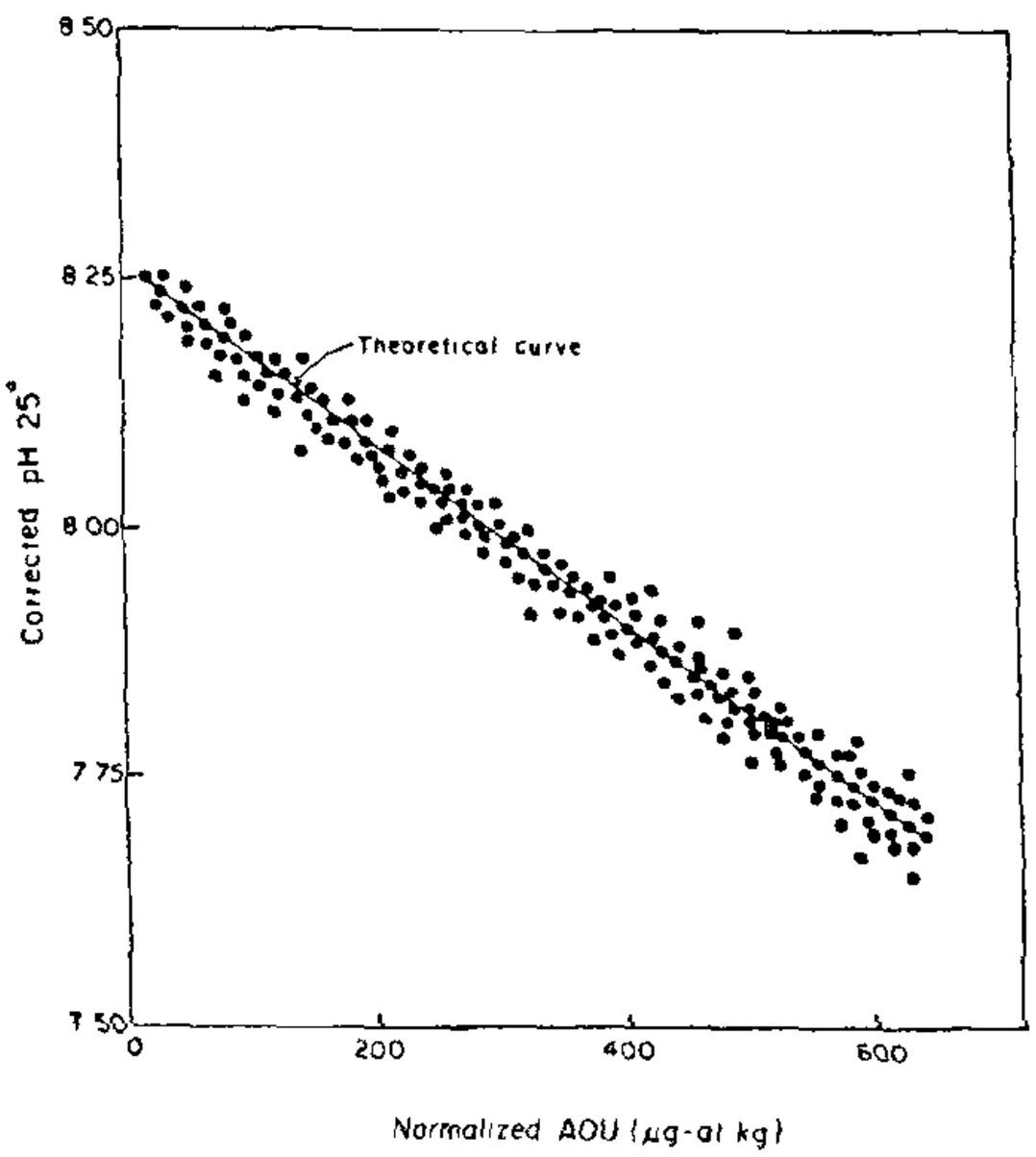

Figure 1. Corrected pH_{25} vs normalised AOU.

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EFFECT OF STREPTOMYCIN ON THE GROWTH OF *RHIZOBIUM*

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THE assumption that the antibiotic resistant mutants and their wild types behave similarly is not necessarily justified because streptomycin being a bacteriocidal compound, is likely to create many distortions in the morphology as well as the physiology of a bacterium.

Streptomycin kills the bacterial cells by blocking protein synthesis on 70 S ribosomes[1]. Some *Rhizobium* strains are reported to have intrinsic resistance against streptomycin and this is used for ecological studies in legume–*Rhizobium* symbiosis[2]. This physiological variation may also bring about a change in the growth rate of the bacteria. In the present study the growth of the wild types and the resistant mutants of chickpea and pigeonpea rhizobia has been compared by following the increase in the turbidity of the growing cultures.

Rhizobium strains of chickpea (*Cicer arietinum* L) and pigeonpea (*Cajanus cajan* L) were isolated either from the nodules of the respective crop plants or from the peat based cultures of the respective rhizobia. After purification and plant infection test, their intrinsic resistance to streptomycin was examined using different concentrations of the antibiotic. Then five mutants of chickpea and four mutants of pigeonpea rhizobia showing a high resistance (1000 μg/ml) were developed using method given by Danso *et al*[3], to study the comparative growth rate of the resistant mutants and their parent types colorimetrically in the YEM broth devoid of streptomycin. The inoculum of each strain (parent as well as mutant) was prepared by adjusting and keeping the same optical density at the time of inoculation. Two tubes of each, the parent and the mutant were inoculated and incubated at room temperature ($25 \pm 2^{\circ}$ C) on a rotary shaker. The percent transmittance of the growing cultures of chickpea and pigeonpea strains was recorded at 12 and 6 hr interval, respectively at 420 nm till the cultures entered into the stationary phase.

Data showed that all the mutant types grew slowly as compared to their parent types. The difference in the growth rates of the wild types and the mutants of chickpea *Rhizobium* in the initial stage of exponential phase was higher (figure 1). Parent culture of S_1 showed faster growth rate (about twice) as compared to its mutant within 24 hr; and later on, the difference in the growth rate narrowed down. Strain S_2 was markedly different from the strain S_1. Upto 12 hr both the parent and the mutant were growing at the same rate but during the next 12 hr the parent culture showed a sharp increase (about two fold) in the optical density as compared to the mutant. The mutant entered into the stationary phase just after 48 hr whereas the parent did not show the typical stationary phase even after 72 hr. The growth rate of the parent types of both S_3 and S_4 strains was higher than their corresponding mutant types but both the parent and the mutant types grew in the same fashion up to 48 hr. The growth pattern of the strains S_3 and S_4 was almost similar. Strain S_5 exhibited different growth trend; the mutant did not differ much in the growth pattern from its parent type except that the parent grew faster when compared to the mutant.

The growth differences in the parents and their mutants of pigeonpea rhizobia were most conspicuous (figure 2). Strain A_1 did not show the characteristic lag

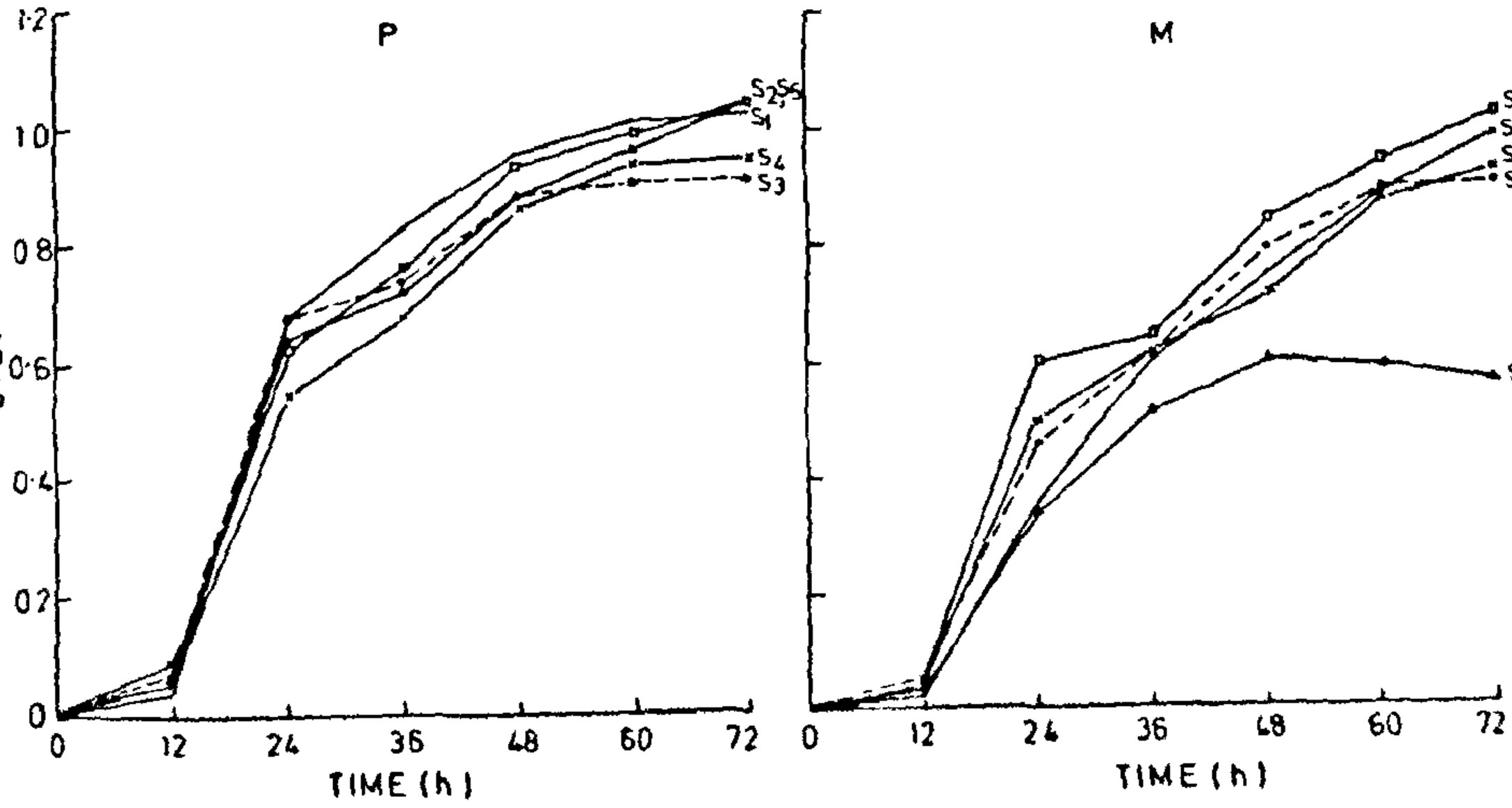

Figure 1. Growth pattern of the parent (P) and the mutant (M) strains of Chickpea *Rhizobium*.

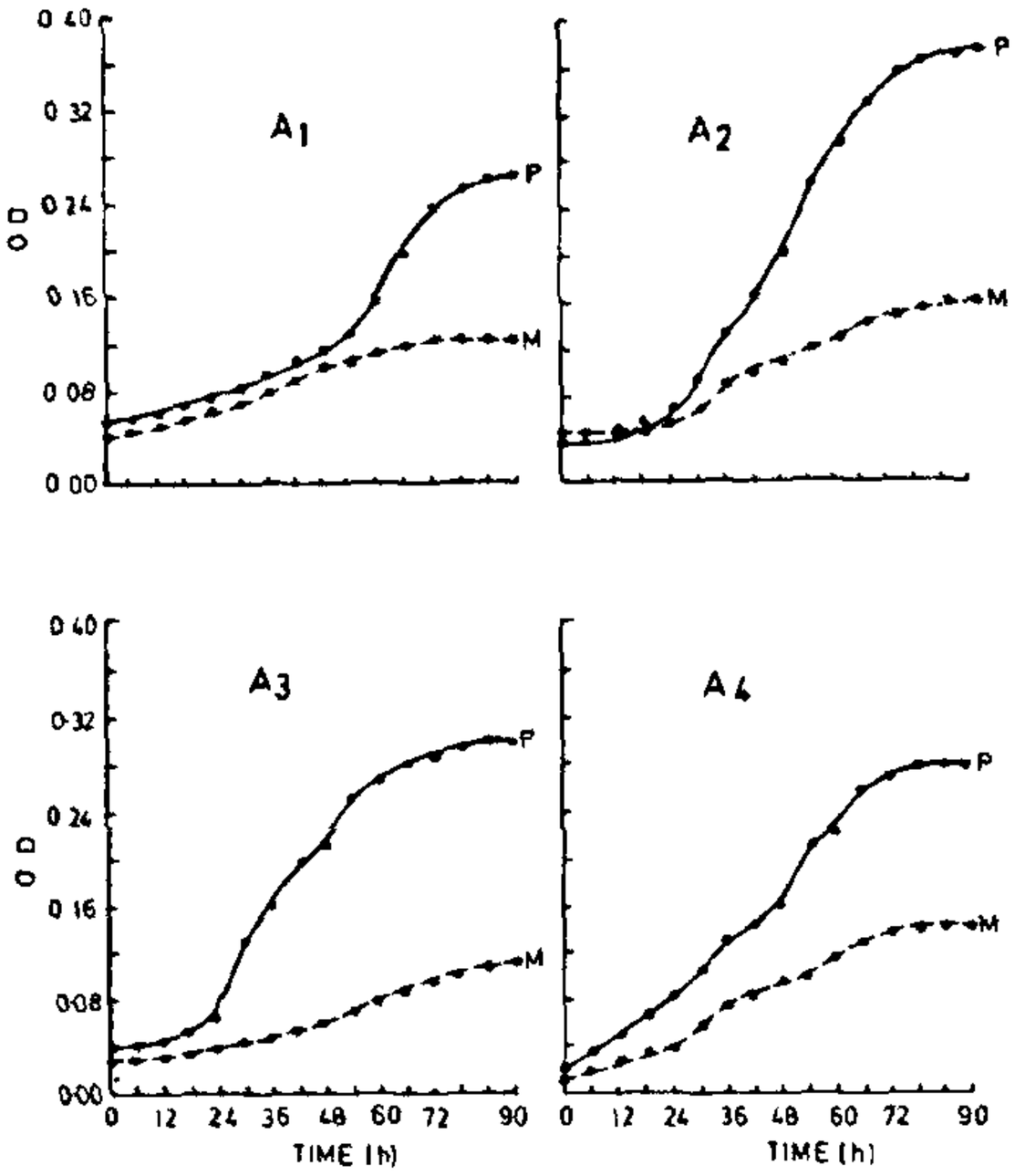

Figure 2. Growth pattern of the parent (P) and the mutant (M) strains of Pigeonpea *Rhizobium*.

phase. The growth pattern of both the parent and the mutant of the A_1 strain up to 54 hr was almost similar; however, later on, the parent culture exhibited a sharp rise in the growth. Strain A_2 showed the characteristic lag phase. The mutant of this strain was relatively slow in growth and showed the lag phase of 18 hr. The maximum difference in the cell density of the mutant and the parent type occurred on the last day of observation. The growth pattern of strain A_3 was almost similar to the strain A_2 and the maximum difference of 173 percent in the optical density of the parent and the mutant cultures was recorded on the final day of observation. Both the mutant and the parent cultures of A_4 did not show the characteristic lag phase. The parent culture of this strain was growing faster than the mutant from the beginning and the difference went on increasing, leading to the highest difference at 78 hr of growth. Pigeonpea rhizobia mutants were comparatively slow in growth rate than chickpea rhizobia mutants.

Not much work has been done on the physiology of antibiotic resistant mutants. In general, it has been observed that the streptomycin resistant mutants of bacteria are slow in growth[4]. In contrast, Dadarwal *et al.*[5] reported positive correlation between the growth rate and resistance of *Rhizobium* to penicillin and streptomycin. Zelazna-Kowalska[5] reported a spheroplast formation in *R. trifolii* strain B, after mutation to a high level of streptomycin (1000 μg/ml).

In view of the above results there appears a serious danger in the use of antibiotic resistant mutants in ecological studies because such slow growing mutants may be poor competitors for nodule forming sites on roots as well as for the other growth limiting factors. In general, a slow growing culture has poor saprophytic competence[7]. On the other hand, many workers[8-10] have reported that the antibiotic resistant mutants of *Rhizobium* are in no way inferior to the parent types in nodulation and other symbiotic properties. To resolve this point, more work is required on the growth rate in medium as well as in soil.

5 May 1985; Revised 23 July 1985

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MYCELIOPHTHORA VELLEREA (SACC & SPEG) VAN OORSCHOT: A NEW RECORD FROM INDIA

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MYCELIOPHTHORA is one of the most conspicuous and