

Table 1 Effect of administration of ganglionic extracts on specific activity of arginase ($\mu\text{mol urea/mg protein/h}$) and urea ($\mu\text{mol g wet wt}$) in hepatopancreas of freshwater snail *Pila globosa*

| | Control | Cerebral | Buccal | Pleuropedal | Supra-intestinal | Visceral |
|--|-------------------|--------------------------------|--------------------------------|--------------------------------|--------------------------------|---------------------------------|
| Active snails administered ganglionic extracts from aestivated snails | | | | | | |
| Arginase | 0.032 \pm 0.002 | 0.015 \pm 0.001 (-53.13%) | 0.011 \pm 0.001 (-65.63%) | 0.024 \pm 0.002 (-25.00%) | 0.01 \pm 0.0003 (-68.75%) | 0.009 \pm 0.0004 (-71.88%) |
| Urea | 0.52 \pm 0.046 | 0.25 \pm 0.013 (-51.92%) | 0.16 \pm 0.001 (-69.23%) | 0.39 \pm 0.06 (-25.00%) | 0.11 \pm 0.008 (-78.85%) | 0.13 \pm 0.009 (-75.00%) |
| Aestivating snails administered ganglionic extracts from active snails | | | | | | |
| Arginase | 0.052 \pm 0.004 | 0.035 \pm 0.002 (-32.69%) | 0.030 \pm 0.002 (-42.31%) | 0.045 \pm 0.003 (-13.46%) | 0.026 \pm 0.001 (-50.00%) | 0.021 \pm 0.001 (-59.62%) |
| Urea | 0.96 \pm 0.071 | 0.52 \pm 0.032 (-45.83%) | 0.36 \pm 0.025 (-62.50%) | 0.76 \pm 0.043 (-20.83%) | 0.25 \pm 0.021 (-73.96%) | 0.29 \pm 0.024 (-69.79%) |

Each value is mean \pm SD of six individual observations.

All values are significant at $P < 0.05$.

arginase² and urea³. Protein was estimated by the method of Lowry *et al.*⁴

The specific activity of arginase and level of urea in hepatopancreas of normal, aestivated and ganglionic extract-administered snails are presented in table 1. The results show that during aestivation the specific activity of arginase increased by 62% and was accompanied by an increase in urea. However, when ganglionic extracts were administered to the snails (extracts from active snails to aestivated snails and *vice versa*), arginase activity and urea level decreased. Visceral ganglionic extract produced the greatest effect on arginase and supra-intestinal ganglion extract produced the greatest effect on urea. In general, the effect produced by ganglionic extracts from aestivated snails was more than that of extracts from active snails.

The increased arginase activity and urea in the hepatopancreas of aestivating snail are due to increased availability of arginine through elevated proteolysis⁵. This elevated arginase activity and urea reflect the metabolic centrality of hepatopancreas in combating ammonia stress during aestivation. When ganglionic extracts of aestivated snails were administered to active snails, both arginase activity and urea level decreased. It appears that endocrine factor(s) in ganglionic extracts caused an increase in urease activity, which in turn enhanced the hydrolysis of urea, resulting in the decreased level of urea. In aestivating snails, ganglionic extracts from active snails had a similar effect, causing reduced formation of urea. The variation in the extent of reduction in arginase activity and urea produced by the different ganglionic extracts indicate the presence of more than one humoral agent in the nerve ganglia.

Prima facie, the altered arginase activity pattern indicates the differential ureogenic capacity of the

tissue. Variation in the urea level is an index of the balance between rates of formation and mobilization. The 'high' and 'low' activity of arginase in the hepatopancreas of aestivating and active snails also exemplify such 'balance'. However, low ureogenic capacity as reflected by decreased arginase activity and low urea content in the hepatopancreas of snails administered ganglionic extracts indicates that hepatopancreas may not be an 'important contributor' to urea production.

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EFFECT OF pH ON PHOSPHATE SOLUBILIZATION BY MICROBES

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THE pH of the nutrient medium is one of the important factors determining the uptake of inorganic

nutrients by plants. The concentration of hydrogen and hydroxyl ions exerts a definite effect on the vital activity of soil microorganisms and also on phosphorus availability¹. The pH of the soil is 5.6–6.2 in sandy soils and 6.0–6.8 in mineral soils². The performance of phosphate-solubilizing microorganisms (PSM) may also be affected if the pH of the nutrient medium is altered. The present investigation was undertaken to evaluate the behaviour of PSM at different pH values.

Fifty ml of Pikovskaya's³ broth was dispensed in 100 ml conical flasks. Tricalcium phosphate (an insoluble inorganic source of phosphate) equivalent to 25 mg P₂O₅ was added separately to each flask. Four sets were prepared, for pH values of 5.4, 6.5, 8.0 and 9.0. The medium was sterilized at 10 psi for 30 min and pH was also recorded after sterilization. The medium was inoculated with 0.5 ml suspension of 4-day-old *Pseudomonas striata*, *Bacillus polymyxa*, *B. circulans*, *B. subtilis* and *Aspergillus niger* cultures (each in triplicate) in Pikovskaya's broth. Uninoculated controls were maintained. Phosphorus was estimated

by the chloromolybdo blue colour⁴ method at 660 nm using Hilger's absorptiometer after 3, 6, 9, 15 and 21 days of incubation at 35°C. Change in pH of filtered medium was also recorded. The sterilization resulted in reduction of the original pH of the medium (from 9.0 to 8.0, 8.0 to 7.0, 6.5 to 6.0 and 5.4 to 5.4), and this is taken as initial pH.

Phosphate solubilization data (figure 1) show that different isolates had maximum dissolution activity at different values of pH. Both incubation period and pH of the nutrient solution were found to influence the performance of PSM.

At pH 7.0 and 8.0, both *P. striata* and *B. polymyxa* showed a gradual increase in phosphate solubilization up to day 9 of incubation followed by a steady decrease. At pH 5.4 and 6.0, however, the increase in phosphate solubilization was recorded up to day 15. Although there was not much variation in the performance of *P. striata* the optimum pH for both these isolates was 6.0.

B. circulans showed increase in phosphate solubilization up to day 15 at pH 5.4, 6.0 and 8.0; at pH 7.0 a

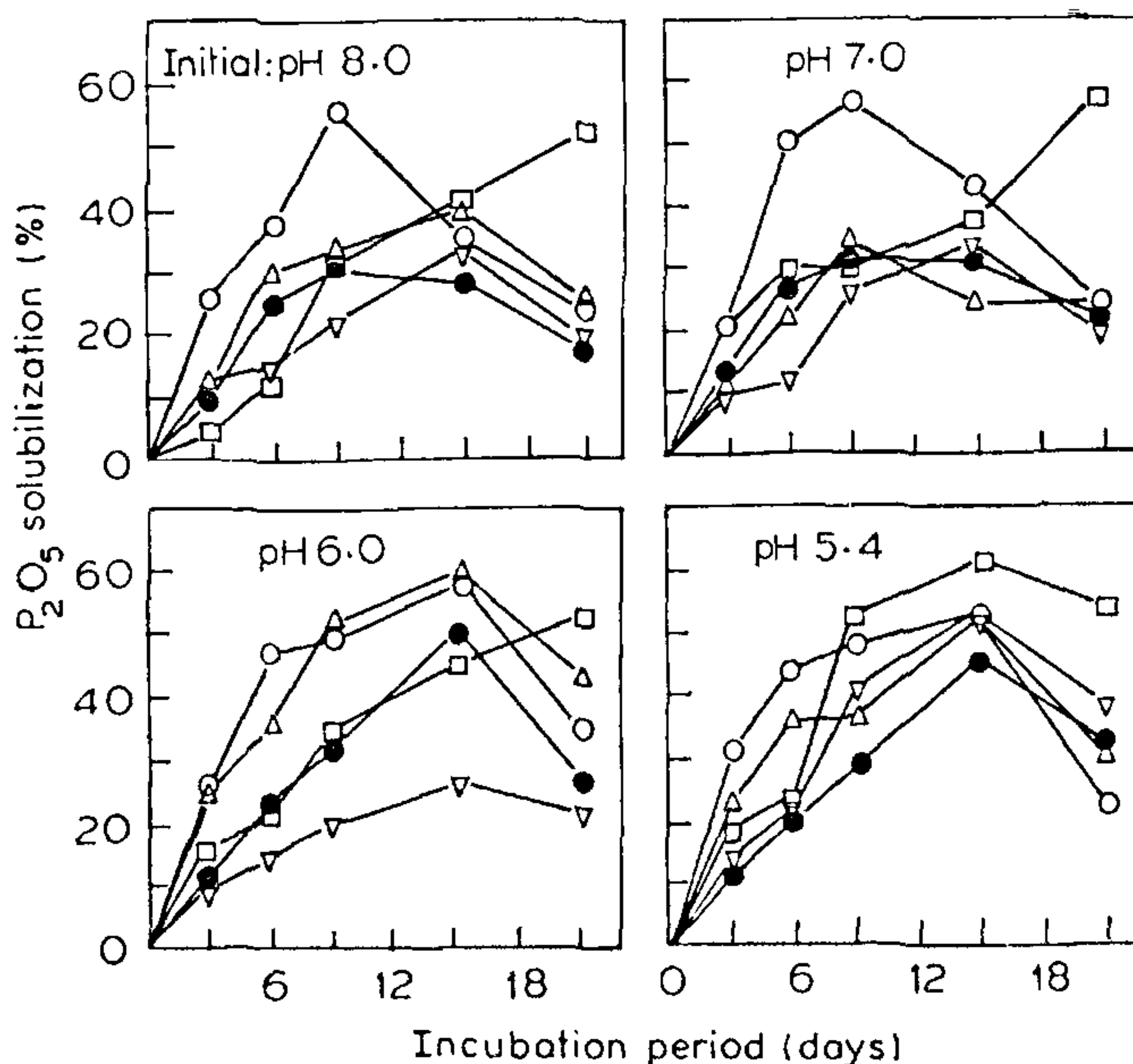


Figure 1. Phosphate solubilization by *P. striata* (○), *B. polymyxa* (●), *B. circulans* (Δ), *B. subtilis* (▽) and *A. niger* (□) at different pH values.

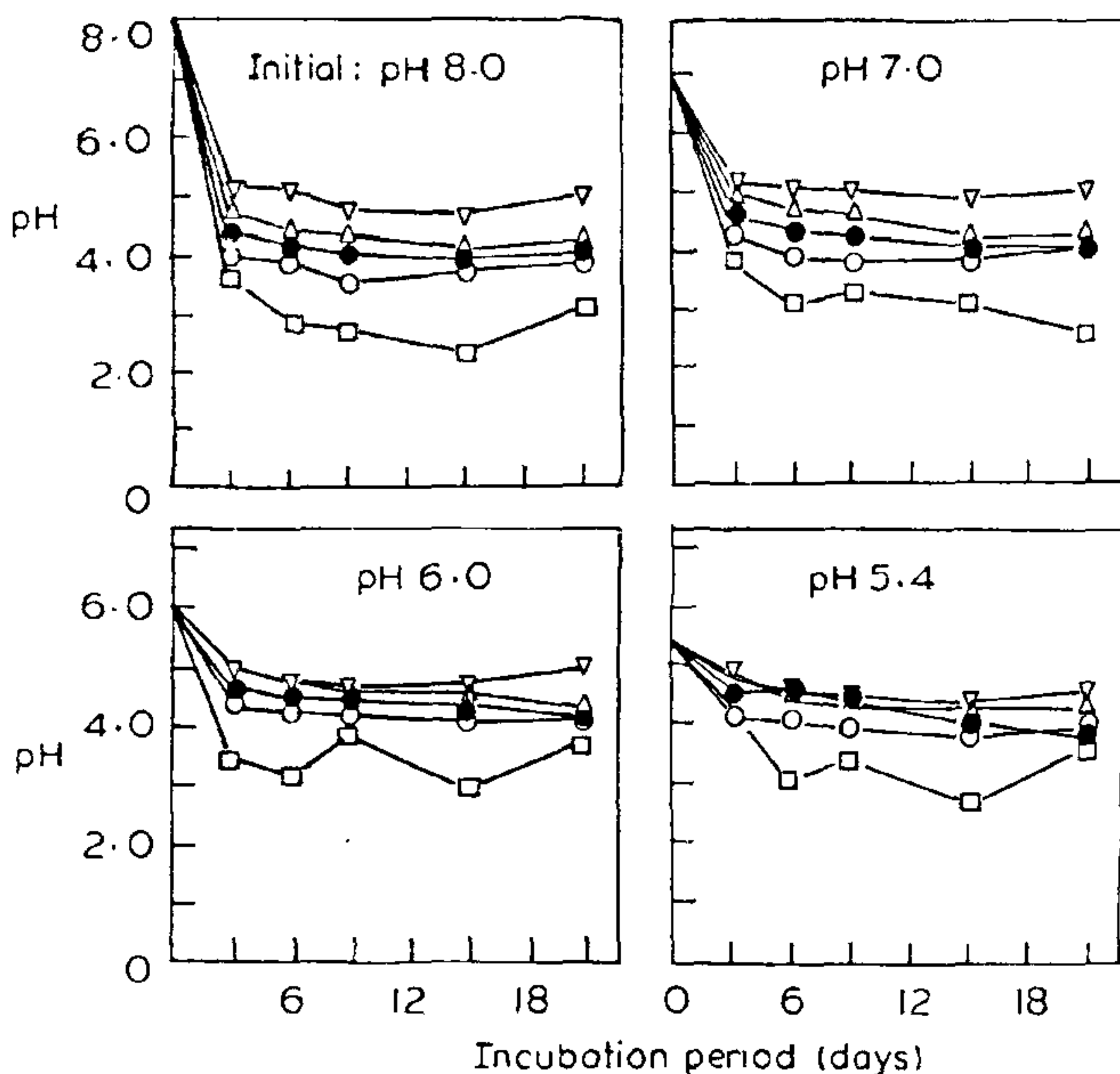


Figure 2. Change in pH of growth medium containing tricalcium phosphate during growth and phosphate solubilization activity of *P. striata* (O), *B. polymyxa* (●), *B. circulans* (Δ), *B. subtilis* (∇) and *A. niger* (□).

decrease was observed after day 9. The pH optimum for *B. circulans* was also 6.0, where the highest dissolution of 60.32% was recorded.

In the case of *B. subtilis* there was an increase in phosphate solubilization up to day 15 at all four pH values, followed by a decrease. However, pH 5.4 was optimum for activity. Thimann⁵ reported the growth of *B. subtilis* within the pH range 4.5–8.5, which supports our findings.

At pH 5.4, the fungus *A. niger* was found to have better solubilization activity than the bacteria *P. striata*, *B. polymyxa*, *B. circulans* and *B. subtilis*. Fungi are known to grow and perform better under medium-acidic condition^{6,7}.

The drop in solubilization after a maximum value may be attributed to deficiency of nutrients in the medium after an optimum period of incubation^{8,9}.

The change in pH of filtered medium (figure 2) was almost in the same range for all four initial values. Drop in pH was highest in the medium inoculated with *A. niger* and least in medium

inoculated with *B. subtilis*. The fall in pH clearly indicates the production of acids, which is considered to be the sole mechanism responsible for the solubilization of insoluble phosphates. There was not much difference in the amount of tricalcium phosphate solubilized between the four organisms.

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HETEROCHROMATIN OF RYE AND TRITICALE—REPETITIVE SEQUENCES

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BANDING techniques have provided valuable information about organizational aspects of chromosomal heterochromatin in rye, wheat and triticales. The chromosomes of the three species show clear structural differences. Rye chromosomes have large blocks of heterochromatin located at the telomeric ends (identifiable by C-banding, figure 1), while in wheat, heterochromatin is present in intercalary positions.

The molecular structure of heterochromatin in rye and wheat has been studied¹⁻³. The studies revealed that the DNA density in heterochromatic regions⁴ is 1.5 to 4 times⁵ higher than that in euchromatic regions. Heterochromatin composed of late-replicating repetitive DNA sequences⁶ has a direct relationship with it⁷. The proportions of single-copy and repetitive DNA in wheat, rye and triticales are presented in table 1. Kinetic analysis (renaturation of denatured DNA) of genome organization in rye and wheat has revealed that repeated sequences are interspersed with unique sequences. The repetitive sequences of heterochromatin of rye and wheat are classified into three subfractions: (i) low repeat (consisting of about ten copies), (ii) intermediate repeat (a hundred to a few thousand copies), and (iii) high repeat (over a million copies). It has also been demonstrated that most of the highly repeated sequences represent constitutive heterochromatin^{8,9} and are species-specific^{10,11}.

The repetitive DNA makes up about 50% of the total DNA in triticales; the highly repeated fraction contains two families, of 3×10^7 and 5×10^6 repeats, which constitute 8.3 and 2.4% of the genome. In rye the repetitive DNA makes up 75-90% of the total DNA and is grouped into four repetitive DNA fami-

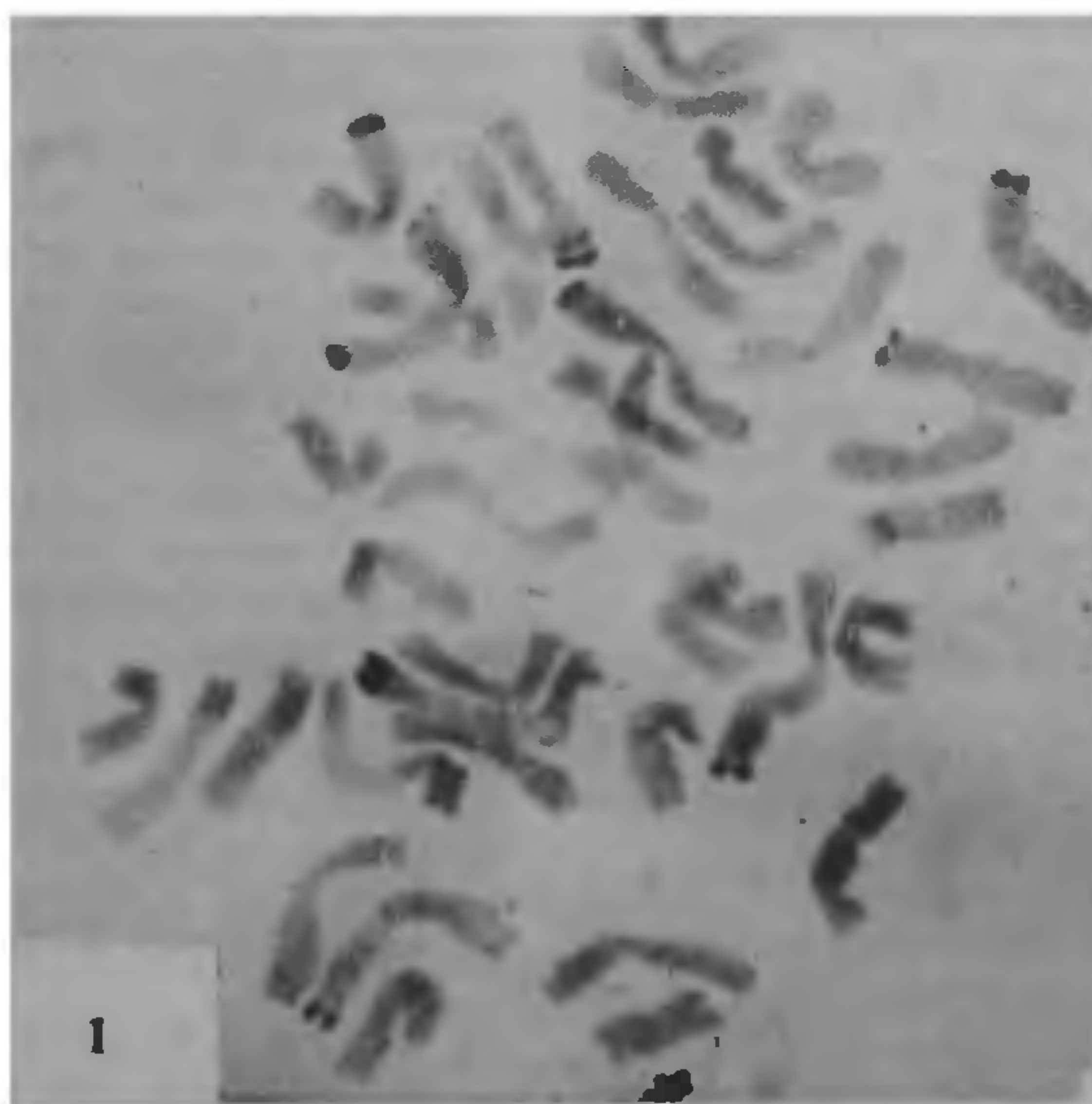


Figure 1. C-banding of rye chromosomes, showing heterochromatin at the telomeric ends.