formation of 3,5-dihydroxyphenylacetic acid, the former was incubated under similar conditions with intestinal micro-organisms, when the formation of 3,5-dihydroxyphenylacetic acid was shown. Administration of 3,4,5-trihydroxyphenylacetic acid to rats resulted in the excretion of 3,5-dihydroxyphenylacetic acid.

To determine whether myricetin breakdown is dependent on the activity of the intestinal microflora *in vivo*, the antibiotic neomycin (100mg) was fed daily to rats before and during the period of myricetin administration, when the formation of 3,5-dihydroxyphenylacetic acid was suppressed.

Incubation of the glycoside myricitrin (myricetin 3-rhamnoside) with intestinal micro-organisms gave rise to free myricetin, 3,4,5-trihydroxyphenylacetic acid and 3,5-dihydroxyphenylacetic acid, indicating that rat intestinal micro-organisms are capable of effecting the cleavage of the glycosidic linkage as well as the heterocyclic ring system of myricitrin.

This work was supported by a grant from the Agricultural Research Council.

Das, N. P. & Griffiths, L. A. (1968). *Biochem. J.* **110**, 449.
Griffiths, L. A. (1964). *Biochem. J.* **92**, 173.

Nakagawa, Y., Shetlar, M. R. & Wender, S. H. (1965). *Biochim. biophys. Acta*, **97**, 233.

The Metabolic Fate of 4-Chloro-2-methylphenoxyacetic Acid in Peas

By D. J. COLLINS and J. K. GAUNT. (*Department of Biochemistry and Soil Science, University College of North Wales, Bangor, Caerns., U.K.*)

This report describes preliminary results from an investigation into the metabolism of the selective herbicide, MCPA,* in plants.

Radioactive MCPA, ³⁶Cl- or carboxy-¹⁴C-labelled, was applied to the leaves of 21-day-old pea plants (*Pisum sativum* var. Progress no. 9). After 24h the plants were extracted with boiling ethanol. Extracts were concentrated and fractionated by partition into ether. Fractions were examined by t.l.c. for labelled metabolites. Two of the breakdown products (MX₁ and MX₂) were found to be ether-soluble, whereas a third was not. In each case both the chlorine atom and the carboxyl carbon remained on the molecule.

This application technique did not permit the use of enough herbicide to isolate sufficient of any metabolite for its chemical identification. Thus incubation experiments with excised plant tissue were set up. Light-grown pea shoot sections were

* Abbreviation: MCPA, 4-chloro-2-methylphenoxyacetic acid.

incubated for 48h in a medium containing 0.5mM-MCPA. Examination of tissue extracts showed the presence of all three previously observed metabolites. Repeated t.l.c. of ether-soluble fractions led to the isolation of products MX₁ and MX₂ as crystalline compounds.

Both substances are non-phenolic acids with u.v. spectra identical with that of MCPA. Compound MX₁ has an i.r. spectrum characteristic of a hydroxylated compound. It was shown to be 4-chloro-2-hydroxymethylphenoxyacetic acid, after synthesis of this compound. Compound MX₂ has an i.r. spectrum characteristic of an amido acid. Acid and alkaline hydrolysis yielded MCPA and a ninhydrin-positive ether-insoluble component. Though still impure, the chemical, chromatographic and spectroscopic properties of compound MX₂ are similar to those of synthetic *N*-(4-chloro-2-methylphenoxyacetyl)-L-aspartic acid.

The ether-insoluble metabolite is also acidic and unstable to hydrolysis. Its purification and chemical characterization are currently under investigation.

D.J.C. is indebted to Ranks Hovis McDougall for financial support. This work was in part supported by grants from the Agricultural Research Council and Imperial Chemical Industries Ltd.

The Microbial Metabolism of Biphenyl

By D. LUNT and W. C. EVANS. (*Department of Biochemistry and Soil Science, University College of North Wales, Bangor, Caerns., U.K.*)

Pure cultures of Gram-negative bacteria were isolated from tropical and temperate soils capable of utilizing biphenyl as sole carbon source in an otherwise mineral salts medium. 2,3-Dihydroxybiphenyl was detected and isolated from the culture; this was cleaved by a particulate fraction from biphenyl-grown cells to give α -hydroxy- β -phenylmuconic semialdehyde [(m.p. 122°C, $\lambda_{\max}$ 350nm (acid) and 430nm (alkali)]. A soluble cell-free extract converted this initial ring-fission product into phenylpyruvate through some, as yet, unidentified intermediates. The subsequent metabolic pathways of phenylpyruvate are known. Unsubstituted biphenyl is therefore hydroxylated in the 2,3-positions, to be followed by a 'meta'-type cleavage between C-3 and C-4 of the aromatic ring; this is to be contrasted with the fate of naphthalene, where a 'meta'-type fission occurs between the angular carbon atom and C-1 of 1,2-dihydroxynaphthalene (Davies & Evans, 1964). Several plant aromatic polymers contain the biphenyl type bond (e.g. the biflavonoids).

Multicomponent mixtures of chlorinated bi-