ERRATA

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The following two communications were printed incorrectly in the issue for June, 1978:
'Paraquat-induced formation of hydroperoxide in mouse liver microsomes' by K.J. Netter & Ch. Steffen
'Metabolism studies with practolol' by D.E. Case, W.E. Lindup, C. Lowery, T.C. Orton, P.R. Reeves & S.E. Whittaker
They are reprinted correctly below.

Paraquat-induced formation of hydroperoxide in mouse liver microsomes

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The activation of oxygen is considered to be the underlying mechanism of paraquat (PQ) toxicity. Bus, Aust & Gibson (1974) demonstrated the formation of superoxide anion and an increased perioxidation of microsomal lipids in a system consisting of NADPH-cytochrome c-reductase, microsomal lipids and a NADPH-regenerating system whereas other authors (Ilett, Stripp, Menard, Reid & Gillette, 1974; Montgomery, 1976) observed a diminished formation of malondialdehyde (MDA).

Mouse liver microsomes were incubated at 37°C in a Soerensen buffer (pH 7.4) with NADPH and a NADPH regenerating system. Oxygen uptake was measured polarographically and MDA formation by the 2-thiobarbitone acid method. Oxygen uptake was increased by PQ in a dose-dependent fashion (K_m 3×10^{-4} M); microsomes from animals pretreated with phenobarbitone had a higher oxygen uptake than microsomes from control animals. MDA formation was decreased by PQ (K_i 6×10^{-5} M) and was not affected by phenobarbitone pretreatment. In the absence of PQ about 1 mole of NADPH was oxidized per mole oxygen. In the presence of PQ (1 mM) the ratio was 2. Addition of NaN_3 (1mM) shifted the $NADPH/O_2$ ratio towards 1 and increased the speed of oxygen consumption in the presence of a NADPH regenerating system. Intraperitoneal injection of 3-amino-1,2,4-triazole which decreases the catalase content in the liver (Heim, Appleman & Pyfrom, 1956) as well as the content of cytochrome P450 (Baron & Tephly, 1969) increased the rate of oxygen consumption as did the addition of azide *in vitro*. Therefore, H_2O_2 seems to be the direct reduction product of oxygen. This is in accordance with the increased oxidation of methanol to formaldehyde in the microsomal system in the presence of PQ which also has been observed by others (Ilett *et al.*, 1974). This agrees with the observed $NADPH/O_2$ ratios.

The effect of PQ on microsomal electron transport is directly comparable to that of menadione which increases NADPH oxidation (Gillette, Brodie & La Du, 1957), O_2 uptake (Sato, Nishibayashi & Omura, 1962) and methanol oxidation with the same $NADPH/O_2$ ratio as PQ does. Menadione, however, is not NADPH-specific. Both substances divert the electrons from the flavoproteins to oxygen, keeping the flavoproteins in the oxidized state. Thereby they inhibit the mixed function oxidations. The inhibition of MDA formation by PQ in this system with a high rate of oxygen reduction does not support the intermediate formation of superoxide anions.

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Metabolism studies with practolol

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A small proportion of patients treated with practolol have been reported to develop adverse reactions termed collectively the oculomucocutaneous syndrome, the occurrence of which does not appear to be related to the pharmacology of practolol (Nicholls, 1976). Practolol metabolism has been re-examined to investigate any abnormality in its disposition in affected subjects; to search for animal species in which extensive metabolism, actually atypical of man, might result in toxic signs in an 'animal model'; and to study the possible formation of metabolites which bind irreversibly to protein.

[^{14}C]-(Acetyl) and [^{14}C]-(phenyl) practolol (100 mg, orally) was given to eight patients, six with skin reactions (Reeves, Case, Felix, Fluke, Holt, Jepson, McCormick, Nicholls & Zacharias, 1978). Peak blood levels (*ca* 1 μg/ml) at 3 h decayed monoexponentially ($T_{\frac{1}{2}}$: 12–16 h). Elimination rates and metabolic patterns were similar in all subjects, 74–90% dose was eliminated in urine, primarily unchanged. Collection of expired air demonstrated 5% deacetylation, although desacetylpractolol was not detected in urine, confirming previous observations (Bodem & Chidsey, 1973). Human adverse reactions do not appear to be associated with gross differences in practolol metabolism.

Of eight animal species studied, mouse, rat and dog most closely resemble man in metabolic profile. Minimal deacetylation (5–14% dose) occurs in all species except marmoset (51%). In other species, practolol was recovered largely unchanged in urine, except hamster, which eliminated practolol primarily as 3-hydroxypractolol and its glucuronide (35% dose).

Microsomal studies (Orton & Lowery, 1977) have been extended to show that hepatic enzymes from several species produce intermediary metabolites which bind to proteins. Hamster microsomes produced the highest binding rate although with marked inter-animal variation (0.44–2.44 nmoles bound mg protein^{-1} 30 min^{-1}). Inhibition and stimulation (53–448% control) were observed in the presence of sodium fluoride, stimulation being the major finding. Tricyclopropene oxide, an epoxide hydratase inhibitor, caused no significant change in binding. *Bis*-[*p*-nitrophenyl] phosphate both inhibited deacetylation *in vitro* and reduced binding. These results suggest, but do not prove, that N-hydroxylation may give rise to the intermediary metabolite(s). Microsomal activation has been utilized immunologically to screen for anti-practolol metabolite antibodies in patients' sera (Amos, Lake & Atkinson, 1977).

The relevance of any hypothesis implicating bound metabolites remains unclear as no toxic signs related to those seen in man were found in albino mouse (18 months, doses up to 100 mg/kg), black (C57 BL/10J) mouse (21 months, 300 mg/kg), rat (24 months, 300 mg/kg), Beagle dog (12 months, 200 mg/kg) and marmoset (6 months, 400 mg/kg). A hamster study reported to us gave rise to no relevant toxic sign. Thus, to date, no animal model for the human adverse reactions is known.

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