Johan Schioldann: History of the Introduction of Lithium into

Medicine and Psychiatry

Birth of modern psychopharmacology 1949

Part II

Renaissance of lithium therapy. Birth of modern psychopharmacology 1949

Chapter 12. Cade's experimental animal studies: the search for the toxic factor. The nitrogenous constituents of urine: urea, uric acid and creatinine

'The first step', Cade[518] explained in 1947, 'was to attempt to devise a method of demonstrating such a toxic agent, if it existed, in the urine of manic persons'. As he had no knowledge of what 'the substance might be, still less anything of its pharmacology for lower animals the best plan seemed to be to spread the net as wide as possible and use the crudest form of biological test as a preliminary investigation'.

For this purpose, Cade set up a primitive laboratory in the pantry of a chronic ward at the Bundoora Hospital, where he single-handedly carried out toxicity tests on guinea pigs.

He started off by testing fresh, concentrated urine from manic patients, and, in way of control, from normal persons, schizophrenics and melancholic patients, injected intraperitoneally into the animals.[519]

Cade observed that the animals generally died in status epilepticus when injected with any sample of concentrated urine, in sufficient quantity, although 'urine from a manic subject often killed much more readily'.[520]

'Now the mode of death is the same, manic and non-manic'; this suggested to Cade that 'the same toxic agent is at work'.[521]

> After a latent period of 12–28 min, in which the animal appears perfectly well, apart from occasional distress of a few minutes following the injection, it becomes tremulous and ataxic, in a few minutes quadriplegic, the hind legs the first affected. It remains fully conscious and often squeals when picked up at this stage. Myoclonic twitching precedes a severe tonic convulsion, and from then on the animal remains unconscious in status epilepticus of tonic type with convulsive gasping until its death a few or many minutes later. An occasional animal recovers.

'To determine the actual toxic agent', Cade decided first to investigate 'the principal nitrogenous constituents of urine',[522] 'the obvious first choices were the end-products of protein metabolism—urea, uric acid and creatinine',[523] and 'it was not very surprising to find that urea was the guilty substance',[524] that 'urea quickly proved to be the culprit'.[525]

Injecting urea solutions in water (4% solution was mentioned) compared with urine, Cade observed 'exactly the same' mode of death, and he made note of the fact that the injected animals either developed no symptoms -'intermediate effects'- or died.[526] Uric acid 0.1% in normal saline solution and creatinine 0.1% in normal saline solution 'appeared to have no toxic effects at all when injected in maximal doses'.[527]

Although Cade thought that 'the actual toxic agent' was identified, he realised that this was 'by no means a complete explanation of what happened when urine was injected, as certain quantitative relationships were far from being fulfilled'. For example, he noted that 'some specimens from manic subjects were lethal in doses of 0.25 to 0.5 millilitre per ounce of body weight', and 'as the lethal dose of 4% urea solution is one millilitre per ounce of body weight, one would have to postulate an impossible concentration of 8% to 16% of urea in these specimens'. Therefore, he said, 'it became necessary to determine what substances modified the toxic effect of urea (the minimal lethal dose), either by diminution or by enhancement'.[528]

For this purpose Cade now compared injections of i) urea 4% solution in water; ii) uric acid 0.1% in normal saline; iii) creatinine 0.1% in normal saline; iv) urea 4% and uric acid 0.1% in water; v) urea 4% with uric acid 0.1% and creatinine 0.1% in water; vi) urea 4% and creatinine 0.25% in water; and finally vii) urea 4% and creatinine 0.2% in water.

The protective action of creatinine

Cade found uric acid to have 'a slightly enhancing effect' on urea's toxic effect (e.g. convulsive fits); however, 'the most surprising result obtained […] was the remarkable protective action of creatinine against the toxic effect of urea'. In fact, it 'more than counterbalanced the mildly enhancing toxic effect of uric acid'.[529]

In view of this 'protective value against the lethal convulsive action of urea on the central nervous system', Cade decided 'to find out' if creatinine had 'any wider anti-convulsant effect'.[530]

In ensuing animal tests, Cade established that creatinine had a 50% reduction of mortality and a 25% complete suppression of convulsive phenomena when injected with Cardiazol and other related agents. He followed this up with a small clinical trial, whose results he found 'though not dramatic were encouraging'. From here, he proceeded to assess 'the protective role' of creatinine in the physiology and pharmacology of the central nervous system. To this end he made some experiments with guinea pigs injected with tetanus toxin and creatinine, only to establish that all animals died, creatinine exerting no protective action. However, he found the likeness between the structural formulae of creatinine and the anticonvulsant Dilantin interesting.

A toxic factor in urine

Cade thought that 'the idea of a toxic factor in urine is not relevant to the use of creatinine as an anticonvulsant, but […] of theoretical importance in the pursuit of the

original investigation'.[531] He added that 'The discovery of the anticonvulsant value of creatinine [was] an unexpected by-product of the original investigation which is still being pursued'.

Cade first thought that 'the extra toxic effect of some specimens of urine from the manic patients might be explicable on the basis of a high excretion level of urea or uric acid, or of a low level of absence of excretion of creatinine'. However, in quantitative urea and creatinine assays (regarding creatinine mentioning Folin, though without reference) he found 'that urine from manic subjects did not differ significantly from other urine in urea or creatinine content'[532] (intriguingly, in this context uric acid was not mentioned). Therefore, he abandoned his assumption that a creatinine-neutralising effect was a sufficient explanation. 'It was more than that', he thought, in that 'the specimens were more toxic than could be explained by the concentrations of urea actually present even if it were being enhanced maximally by uric acid. One would have required a urea concentration of 8% to 16%'.

Therefore, in Cade's opinion, it was 'difficult to avoid postulating a third substance in urine which more than neutralizes the protective action of creatinine against the toxic activity of urine'.

He speculated whether the following factors were 'operative: i) the toxic effect of urea; ii) the protective effect of creatinine; iii) a third toxic substance which a) neutralizes the protective effect of creatinine and b) enhances the toxic effect of urea'.[533]

From here Cade proceeded to postulate that a minimal lethal dose can be established, the urea and creatinine concentration of urine being known, 'on the assumption that no other factors are involved'. He postulated further that the ratio of the calculated minimal lethal dose to the actual minimal lethal dose is a measure of the presence of the third factor. Finally, based on 'bulk specimens of urine from manic subjects', more or less toxic, he stated that this ratio had a general numerical value of two to four, 'that is, the actual minimal lethal dose is two to four times less than the calculated dose'.

November 11, 2021

[518] Cade, 1947, op. cit.

[519] ibid.

[520] ibid. cf. Cade JF.: 'Lithium in psychiatry: historical origins and present position'. Editorial. Aust. NZ. J. Psychiatr. 1967;1:61–62. According to Cade, the urine from some manic patients would kill the guinea pigs in as low a dose as 0.25 ml per 30 gm body weight, whereas the most toxic control specimen was not lethal in doses lower than 0.75 ml per gm body weight. He concluded therefore that 'all that had been demonstrated so far was that any concentrated urine in sufficient quantity would kill a guinea pig, but that urine from a manic subject often killed much more readily'.

[521] Cade JF.: 'Lithium—past, present and future', in: Johnson FN, Johnson S.: 'Lithium in medical practice. Proceedings of the First British Lithium Congress, University of Lancaster, England. 15–19 July 1977'. Lancaster: MTP Press, 1978. pp.5–16. cf. Cade 1967, op. cit.

[522] ibid.

[523] Cade JF.: ‘Lithium in psychiatry: historical origins and present position’. Editorial. Aust. NZ. J. Psychiatr. 1967;1:61–62.

[524] Cade, 1947, op. cit.

[525] Cade, 1967, op. cit. 1:61–62.

[526] Cade, 1947, op. cit.

[527] ibid.

[528] ibid.

[529] ibid.

[530] ibid.

[531] ibid.

[532] ibid.

[533] ibid.