On the discovery of UREA. Identification, synthesis and observations that let to establishing the first uraemic retention solute

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Although the discovery of UREA in urine is frequently attributed to Hermann Boerhaave **[1]**, from Leiden, the first description we know of this particular urinary salt is from Jean Baptiste Van Helmont. He described a salt that "never occurs outside man's body", that "differs from sea-salt, also present in urine, by remaining unchanged in its course through the body and on putrefaction of urine", and added "the sea-salt *in its cooling, adheres to a wooden vessel even while it is separated from saltpeter, but the salt of urine grows together in the bottom of the liquor"* **[2]**. Jean Baptiste van Helmont (1577-1644), was a Brussels born chemist and physician, founder of the iatrochemical school which looked to chemical explanations of vital phenomena. He was a man of great intellectual curiosity and studied philosophy at Louvain.

The best known text on the description of urea and also on the method for its purification is from Hermann Boerhaave [1] (1668 – 1738), a Leiden born botanist and chemist who greatly participated in the renown of the Leiden University during the XVIIth and XVIIIth century. He wrote in his "Elementa Chemiae":

"Take some very fresh well-concocted Urine of persons in perfect Health, put it preferently into a very clean Vessel, and with an equable Heat of 200 degrees, evaporate it till you have reduced it to the consistence of fresh Cream" …"Put a large quantity of this thick inspissated Liquor into a tall cylindrical glass vessel with a paper tied over it and let it stand quite in a cool place for the space of a year..."

"By this means, then, you will have a solid, hard, sub-pellucid, brown saline mass, fixed all about the bottom of the Vessel; and over this, a thick, black, pinguious liquid, separated and rejected as it were from the concreted Salt …"

"Decant, take out the saline mass, put it into another Vessel, pour some very cold water upon it and shake it about to free it from its oily Impurities which may be done pretty easily, as it will not readily dissolve in cold Water…"

"Keep this saline matter under its proper title.

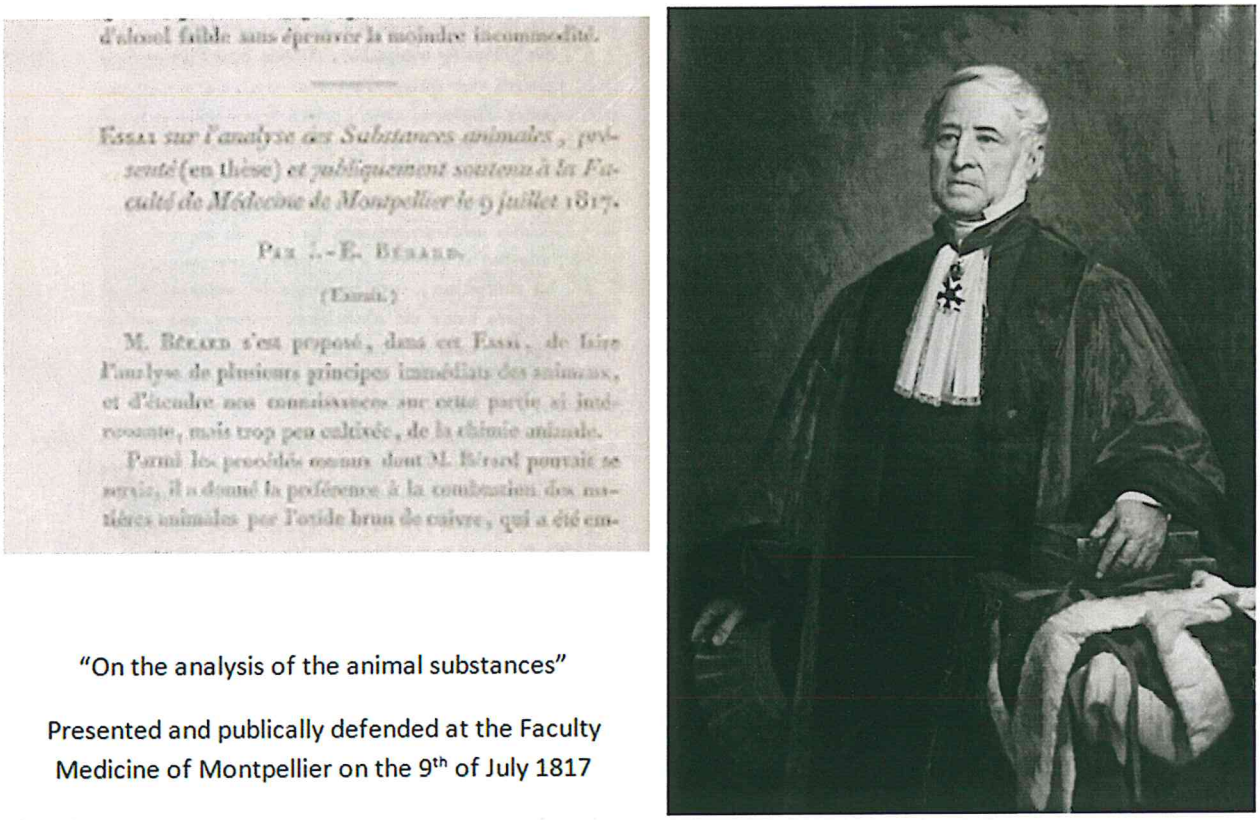
If this is dissolved in hot Water, and strained till the Lixivium becomes exceeding limpid, and evaporated to a Pellicle in a very clean glass Vessel, then, if you set it by a cold place, it will shoot into saline Glebes of a particular kind, that are perfectly distinct from every other Salt.

In their figure, and solidity, however, they come pretty near to the Crystals of sugar. These are not fetid, nor alkaline but very volatile.This is the native Salt of urine."

Boerhaave described in detail how urea was indeed in urine (an intrinsic element of a living animal or human being). He already proposed that to obtain urea is preferable to use someone normal and described a method

which included steps longer than 1 year, as he left the crude material he obtained urea from for one year in a cool place before he used it.

Fourcroy and Vauquelin **[3]** in 1808, gave this substance the name of "*urée*" because it was specific of urine, a product clearly specific of living animals and Bérard in Montpellier in 1817**[4]**, completed the remarkable description of the chemical composition of urea by William Prout in London **[5]**, following the work by Fourcroy and Vauquelin in France. Jean – Etienne Bérard (Figure 1), reported that the composition of Urea from urine contained 43.4 % of Nitrogen, 19.4% of Carbon, 10.8% Hydrogen, and 26.4% Oxygen [4].



d'alcool faible sans éprouver la moindre incommodité.

Essai sur l'analyse des Substances animales, présenté (en thèse) et publiquement soutenu à la Faculté de Médecine de Montpellier le 9 juillet 1817.

Par J.-E. Bérard.

(Extrait.)

M. Bérard s'est proposé, dans cet Essai, de faire l'analyse de plusieurs principes immédiats des animaux, et d'étendre nos connaissances sur cette partie si intéressante, mais trop peu cultivée, de la chimie animale.

Parmi les procédés connus dont M. Bérard pouvait se servir, il a donné la préférence à la combustion des matières animales par l'oxide brun de cuivre, qui a été em-

"On the analysis of the animal substances"

Presented and publically defended at the Faculty Medicine of Montpellier on the 9th of July 1817

Figura 1 di 1 Jacques – Etienne Bérard, AnnalChim. Phys., 5, 290 (1817) Acknowledgment : Courtesy of Université Montpellier, U.F.R. de médecine. BIU de Montpellier. Service photographique.

Pursuing the work contributed by the chemists, Friederich Wöhler dramatically changed the view of this substance that was different from sea salt and thought to be exclusively produced by animals when he wrote to Berzelius in Stockholm: "*I can make urea without needing a kidney, whether of man or dog. The ammonium salt of cyanic acid is urea*" ($HCON + NH_3 \rightarrow H_2N\text{-}CO\text{-}NH_2$). Wöhler had succeeded in the synthesis of urea outside from an animal and established the starting point of modern organic chemistry. Interestingly, Wöhler was upset by his discovery. He wrote to Berzelius, *"The great tragedy of science, the slaying of a beautiful hypothesis by an ugly fact."* Vitalism, which proposed that the compounds of the living organisms were unique, submitted to the *"life's spark"*and could not be made from inorganic materials, was the dominant thinking in that time in Europe. Wöhler certainly also adhered to this theory and his discovery making an organic compound from inorganic materials was contradicting his own believes.

Physicians greatly contributed to the understanding of the production of urine and urea excretion. Joseph Nicolas Comhaire (1778-1860) observed that when both kidneys are removed from a dog, no urine is accumulated in the bladder, showing that the kidneys are at the origin of the urine **[6]**.

In 1821, Prevost (1790-1850) and Dumas (1800-1884) reported their experiments with dogs that they nephrectomised in the Society of Physics and Natural History in Genève in 1821 **[7]**. They first explained their observations on dogs:

"When one is to look the physiologic phenomena following the removal of the kidneys, it is preferable to first remove the right kidney, since its connections with the liver, and leave a fifteen days delay between this procedure and the next one. The first one, if it has been well performed does not alter the health of the animal…", "...When the animal has lost its second kidney is merely affected before the third day…", "…Finally, all the mentioned symptoms worsen, weakness increases, and the animal dies between the days 5 and 9. If one removes both kidneys at once, the resulting inflammation shortens this time lapse, and the subject seldom goes until days 4 or 5..."

Then they studied the blood and the urine of these animals and concluded that the urea observed in blood was the same as the urea found in urine and indeed, when dogs were binephrectomised doubled the amount of urea in blood: « We have observed that the same procedures on the blood of the binephrectomised animals produced twice as much alcohol residue »… « giving a white and crystalline substance which was entirely soluble in water » which they analysed and compared to the composition reported by Bérard and they concluded as follows: « The difference merges with the errors possible in this type of analyses, and we think that it is permitted to conclude that the urea from the blood and that of urine are identical[7]»

This work set the basis for the "humoral" view of renal physiology, by opposition to "morbid anatomy" theory, supported by Bright and which was dominant in that time in Europe (early 1800). Further work in refining Liebig's method to dose urea contributed by Joseph Picard (1834-1896) **[8]** enabled him to see a negative gradient between renal artery and vein: the kidney removed urea, whilst a positive gradient was observed between carotid artery and jugular vein showing that the brain did not. The basis for renal physiology was set.

Friedrich Th. von Frerichs (1819-85) introduced in 1851 the term "Uraemia" and the concept of retention solute when commenting on Bright's reports **[9]**, and R. Christison **[10]** and JC. Gregory**[11]** introduced the putative toxic effect of uraemic retention solutes. Urea has been since, the most frequently used compound to assess kidney diseases and it has given the name to kidney related dysfunction: "uraemia" and uraemic syndrome are what we know presently as chronic kidney disease (CKD), and as such urea deserves a prominent place in the list of compounds known to be altered during renal disease [12]. Urea has not only been used to assess renal dysfunction, but is the most widely used parameter to assess the quality of replacement therapy, although the toxicity of urea is still under debate more than 200 years after its discovery [13].

Addendum

The description of the discovery of urea is very nicely reported by G Richet in the HISTORICAL ARCHIVES series of *Kidney International* [14]; very much advised reading.

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