

SYNTHESIS AND COMPARATIVE ANALYSIS OF PHARMACEUTICAL FORMULATION OF PARACETAMOL

Musa N. U.* , Ambi A. A., Usman A. Y., Hassan A., Umar A., Mustapha H.G.

¹Department of Science Laboratory Technology
Federal Polytechnic, Bauchi P.M.B 0231 Bauchi

*Correspondence Author Email: mnuraumar@fptb.edu.ng

Abstract

Paracetamol was synthesized by nitrating phenol using sulfuric acid and sodium nitrate. The para-isomer was separated from the ortho-isomer by fractional distillation. The 4-nitrophenol was reduced to 4-aminophenol by a reducing agent. The 4-aminophenol was reacted with acetic anhydride and paracetamol was obtained. Percentage of active ingredients, spectrophotometric and their absorption bands at 257nm were analyzed on eight different brands of commercially compounded paracetamol. Six different brands were found to contain up to standard percentage of active ingredients of (90 to 100%) according to British pharmacopeia, while only two brands contain low percentage active ingredients (i.e. below 90%).

Keywords: - C₆H₅NH₂- Aniline, BH₄ – borohydride, ABCDEFGH – commercially produced paracetamol samples, antipyretic, acetanilide, phenacetin, p-aminophenol, nitration, acetylation and *N*-(4-Ethoxyphenyl) ethanamide- paracetamol.

Introduction

Drugs are chemical substances that take effect on functioning of living thing and the organisms (such as bacteria, fungi and protozoans) that infect them. Pharmacology the sciences of the drugs, deals with all aspect drugs in medicine, including their mechanism of action, physical and chemical properties, metabolisms therapeutics and prophylaxis.

N-(4-hydroxyphenyl) ethanamide, otherwise known as Paracetamol or acetaminophen depending on where you live in the world, is one of the most widely used over the counter drugs. It has the molecular formula $C_8H_9NO_2$. It is an analgesic (pain reliever) and also an antipyretic (fever reliever). For these reasons it is used to relieve a person of mild to moderate pain, for example; toothache, headaches or symptoms of a cold and to control fever (high temperature, also known as pyrexia). For pain relief it works by interfering with certain chemicals in the body called prostaglandins. Prostaglandins were first discovered in the 1930's from human semen, thinking the chemicals had come from the prostate gland he named them prostaglandins, but it's since been established they are synthesized in every cell in the body. They act as chemical messengers like hormones but do not move to other sites, they stay in the cell that they were synthesized in. Prostaglandins have a variety of physiological effects, one being that they are released in response to pain or injury, paracetamol works by inhibiting the production of prostaglandins making the body less aware of the pain or injury. Paracetamol reduces temperature by acting on an area of the brain called the hypothalamus, responsible for regulating body temperature.

Until the mid-19th century, the approach to drugs therapeutics was entirely empirical. This thinking changed when the mechanisms of drugs action began to be analyzed in physiological terms and when some of the first chemical analysis of naturally occurring drugs was performed. The end of the 19th century signaled the growth of the pharmaceutical industry and the production of the first synthetic drug. Chemical synthesis has become the most important source of therapeutic drug (Peter, 1995)

The history of paracetamol is an interesting one, at the approach of the 20th century, the discovery and synthesis of medicines was rather arbitrary, with scientists generally just testing new compounds on humans straight away and then observing if it had positive (or negative) effects. The story of paracetamol starts with the first aniline (also known as phenylamine or aminobenzene) derivative to be found to possess analgesic and antipyretic properties, acetanilide.

Aniline

Aniline is an organic compound with the molecular formula $C_6H_5NH_2$, shown above, consists of a phenyl group attached to an amino group. The new potential medicine acetanilide had been synthesized simply by the aniline gaining a secondary amide group, by reacting the aniline with ethanoic anhydride, ethanoic acid would also be produced. The reaction is shown below.



Acetanilide

The discovery was soon published and acetanilide medication was soon in production in 1886, remaining in use for several years due to how cheap it was to produce. But although acetanilide was shown to act as being effective in reducing fever and relieving mild pain, a search for less toxic aniline derivatives started because of some of the awful side effects acetanilide had, for instance cyanosis (appearance of blue or purple coloration of the skin due to tissues near the skin being low in oxygen) caused by it deactivating haemoglobin in erythrocytes.

The search led to a new derivative that was antipyretic and analgesic and was less toxic than acetanilide called N-(4-Ethoxyphenyl) ethanamide. Marketed in 1887 under the name phenacetin, it has remained in use ever since but has declined in its use due to its adverse effects on the liver. It has the chemical formula $C_{10}H_{13}NO_2$.

N-(4-Ethoxyphenyl)ethanamide

In 1893 Joseph von Mering improved on phenacetin producing paracetamol, but mistakenly thought it had the same adverse effects as acetanilide. In the 1940's it was realized that paracetamol was a major metabolite of phenacetin; it was then considered to quite possibly be the component that caused phenacetin to have the desired effects and that the negative effects were caused by a minor metabolite released. Then in 1953 paracetamol hit the markets, being promoted as superior to aspirin in that it was safe for children and with people with ulcers. (UKEssay.com 2018)

Scope

- i. The study aimed at synthesizing Paracetamol by nitrating phenol using acid in the presence of sulphuric acid. The para-isomer will be separated from the ortho-isomer using distillation apparatus. The product will be reduced to 4-aminophenol using a reducing agent in basic medium. The 4-aminophenol will be reacted with acetic anhydride to give paracetamol.

- ii. To calculate the percentage content of paracetamol from the commercial tablets and compare the purity to the synthesized sample
- iii. Physical analysis will be carried out on this product as well as other commercial products of paracetamol.

Sources of material

Different brands of paracetamol tablets were purchased from various pharmacies within Bauchi metropolis, Bauchi state - Nigeria . Other reagents and apparatus were obtained from chemistry laboratory of the department of Science Laboratory Technology of the Federal Polytechnic Bauchi. Spectrophotometric analysis was carried out at chemistry laboratory of Abubakar Tafawa Balewa University Bauchi.

Synthesis of paracetamol

Phenol is nitrated in the following ways;

Chemical Method

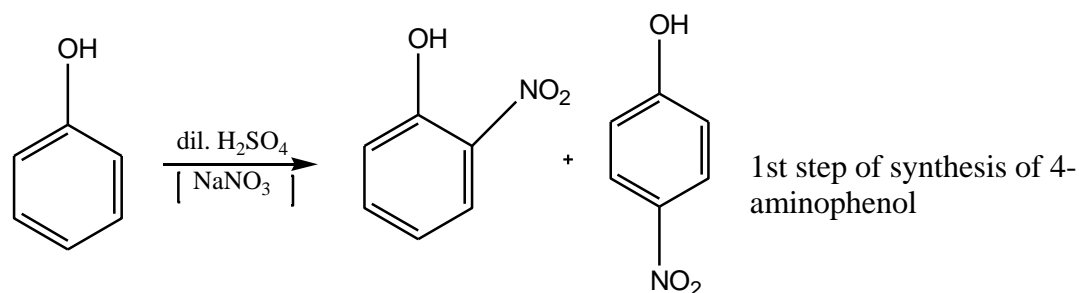
The synthesis of the amide essentially just requires running the reaction under certain temperature condition with an appropriate catalyst. Paracetamol is prepared from p-aminophenol by acetylating it with acetic anhydride in the presence of 3-4 drops of concentrated sulphuric acid as catalyst.

Nitration of phenol

The three reagents were thoroughly mixed and the flask was fitted with a reflux condenser.

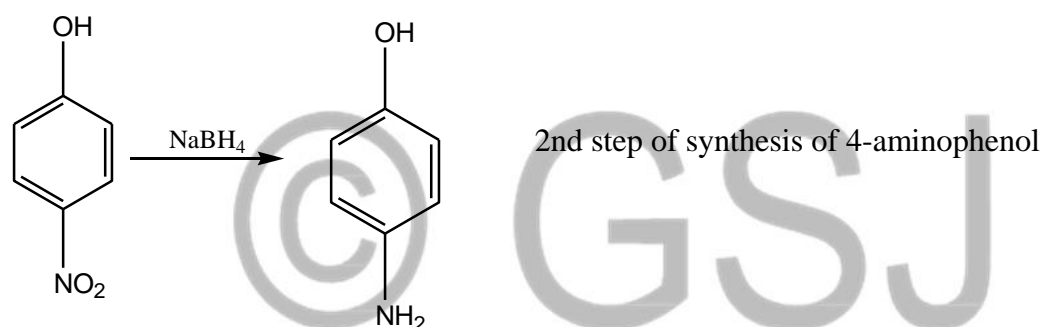
The temperature was raised to 60⁰c for some time. The apparatus was allowed to cool down and the content of the flask was poured into 75cm³ of cold distilled water in a beaker. The water layer was washed with 75cm³ of sodium carbonate (50% Na₂CO₃) solution, and finally with 75cm³ of distilled water again.

4-aminophenol, the building block of paracetamol (reacting 4-aminophenol with ethanoic anhydride give paracetamol) which is a primary amine. 4-aminophenol is made by reacting phenol with sulphuric acid and sodium nitrite which gives two products, 1-nitrophenol and 2-nitrophenol, 2-nitrophenol is then reacted with sodium borohydride that give rise to 4-aminophenol.



Reduction of Para-nitrophenol

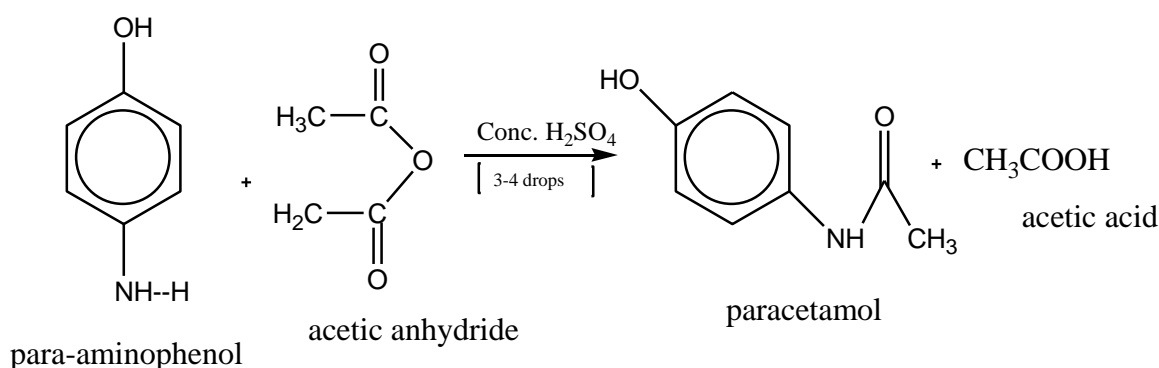
A solution of 7.0g (0.05mol) of p-nitro phenol was added in 250ml of 2M sodium hydroxide, drop wise in a separating funnel, within few minutes. It was stirred at room temperature until the yellow colour disappeared (about 10 min) and was filtered. The filtrate was acidified with 2M HCl to destroy excess borohydride and was neutralized with dil. NaOH. The product was extracted with 450ml portions of ether and evaporated.



P-aminophenol was obtained as white solid melting point 167-69°C as Wiyid nulls absorbs at 3350-3300cm.

Acylation of P-aminophenol

About 11g (0.1mol) of p-aminophenol was suspended in 30ml of water contained in a 250ml beaker. 12ml (0.1mol) of acetic anhydride was warmed with the mixture, stirred vigorously and warmed on a water bath. The solid dissolves after 10 minutes it was cooled, filtered. The solid acetyl derivative at the pump and washed with little cold water. It was recrystallized from hot water and dried upon filter paper in the air. The yield was Para acetyl aminophenol is 169 and 93%.(Graig Barter, 2005).



Determination of relative density

The procedure employed was described by Anthony (2007). Two density bottles were washed allowed to cool down in a desiccator. Each bottle them was filtered with water and the other with were taken and recovered.

The sample density was determined by the difference in weight.

Relative density = weight of simple/weight of an equal volume of water

$$= \frac{W_2 - W_1}{W_4 - W_3}$$

Where W₁ = weight of empty bottle A

W₂ = weight of empty bottle A + sample

W₃ = weight of empty bottle B

W₄ = weight of empty bottle B + water

Melting point determination

Melting point apparatus was employed to determine the melting point of the sample capillary tube was scaled with the help of bunsen flame and it was filled with the sample and inserted on the machine. Temperature was recovered with a thermometer.

Identification

- i. Melting point is about 169°C.
- ii. Reaction of paracetamol with HCl: 0.1g of the sample was boiled with 1ml of conc. HCl for 3 minutes. 10ml of water added and cooked with no precipitate will be observed. (Graig Barter, 2005).

Assay of Paracetamol in commercial tablets (Spectrophotometric method)

- i. Ten tablets were weighed and powdered.
- ii. A quantity of the tablet equivalent to 180mg of paracetamol (calculations see appendix) was added to 50ml of 0.1M NaOH in 250ml volumetric flask.
- iii. The solution was diluted with 100ml of distilled water and was shaken on a magnetic shaker for 15 minutes at 200rev/min. it was mark with distilled water.
- iv. 10ml were filtered and diluted with 90ml of distilled water.
- v. 10ml of the resulting solution was added to 10ml of 0.1M NaOH and diluted with 80ml of distilled water.
- vi. The absorbance of the resulting solution was taken at 257nm using 0.1M NaOH as the blank.
- vii. The content or concentration taken 715 as the value of A (1%:1cm³) at the maximum of 257nm.

Experimental results

Physical analysis

Table 1: Physical Assessment of Paracetamol

<i>Sample</i>	<i>Density (g/cm³)</i>	<i>Ph</i>	<i>Melting point (°c)</i>
A	1.2624	5.5	169
B	1.2428	6.0	165
C	1.2260	6.1	167
D	1.2623	7.4	170
E	1.2622	8.0	168
F	1.2630	7.5	169
G	1.2631	8.1	168
H	1.2633	6.5	168
X	1.2630	7.4	169
Range	1.2600 to 1.2633	4 to 9	168 to 174

Musa N. U 2020

Reaction with HCL

- a) Visible reaction = no precipitate observed
- b) Inference = presence of paracetamol

Comparative instrumental analysis of commercial pharmaceutical formulation of Paracetamol

Table 2: Spectrophotometric Assay

<i>Sample</i>	<i>Av. Weight (g)</i>	<i>Equ.weight (g)</i>	<i>Absorbance at 257nm</i>	<i>Percentage recovery</i>
A	0.5487	0.1975	0.4736	92.00
B	0.5849	0.2105	0.4424	85.96
C	0.5831	0.2099	0.4576	88.80
D	0.5404	0.1945	0.4704	91.30
E	0.5600	0.2016	0.4854	94.40
F	0.5695	0.2050	0.4904	95.20
G	0.5493	0.1977	0.4928	95.70
H	0.5530	0.1990	0.4728	91.80
X	0.6605	0.1902	0.5070	98.48

Musa N.U 2020

Normal range for percentage recovery, 90-100%

Discussion

Based on the results obtained from table 1. Nine samples were analyzed samples A, B, C, D, E, F, G, H and X were found to have densities which lies on the range of 1.2260 to 1.2633. This range 1.2260 to 1.2633 was the referenced range of densities for paracetamol at NAFDAC aerial laboratory Kaduna as stated in British pharmacopeia 2004.

Samples A, B, C and H were tested to have acidic properties because of their low pH values. Samples E and G have alkaline properties while samples D, F and X have a neutral pH values. Referring to the objective of the study is to produce a pain reliever which is free from acidic properties, the synthesized compound X was found to have a

neutral pH, likewise samples D and F. This means that they have no acidic or alkaline properties. Other samples (E and G) are also free from acidic properties and the rest samples A, B, C and H have acidic properties. According to Peter 1995, "It has long been thought that gastric and duodenal ulcers are related to the production of acid, excess level of this acid together with the enzyme pepsin quickly produces ulceration unless the stomach is protected". Introducing this acidic drug to ulcer patients will increase the degree of ulceration

While the melting points of all the compounds were found to fall within the normal range of 168-170°C This is the standard range of NAFDAC aerial laboratory Kaduna as stated in British pharmacopeia 2004.

In table 2, all the samples were tested for their absorbance with spectrophotometer. Seven samples A, D, E, F, G, H and X was found to contain up to standard percentage active ingredients which falls between the ranges of 90-100% according to British pharmacopeia 2004. While samples B and C have lower percentage active ingredients. This low value will reflect on the required analgesic properties of the drug as it affects some of the physical properties.

All the samples were tested to have a required weight which ranges from 0.4800grams to 0.6800grams according to British pharmacopeia 2004.

Conclusion

Based on the study it can be concluded that not all commercially compounded paracetamol contains the required percentage active ingredient as in this case of samples B and C, they have the required weight but their absorbance was as required which may attribute to other physical properties such as low pH values etc. Acidic paracetamol is very dangerous to ulcer patient as stated earlier in the literature by Peter 1995 earlier. Analysis should be expanded by such industries that produces such type of drugs to reveal their acidity and should be stated clearly on the label, perhaps pharmacist will be guided when administering such drugs to the patients.

Recommendation

Based on the research topic, the aim of producing an analgesic free from acidic property was successfully carried out. The following recommendations were made for further findings.

- i. The effect of interacting variables on the compaction performance of paracetamol with different starch tablet should be carried out to study which starch is good for compaction.
- ii. Effect of additives like colorants, sweeteners e.t.c should also be studied.
- iii. Further recommendation goes to the relevant authorities that they should enhance and equip regulatory agencies such as NAFDAC, SON etc. with laboratory apparatus to extend their work to various state capitals so that they will analyze the drugs that are available in the market to reduce their adverse risk or abuse.

References

- [1] Anthony S. Travis (2007) "*manufacture and Use of Anilines*": 'A vast Array of processes and product'. **The Chemistry of Anilines** part : 765.
- [2] Daly F.F, Fountain J.S, Murray, Gaudiness A.& Buckley N.A (2008) "Guideline for the management of paracetamol poisoning in Australiaand New-Zeland Explanation and Elaboration". A consensus statement from Clinical Toxicologist Consulting to the Australian Poisons Information Center. **Medical Journal Australia** **188**(5): 296-301.
- [3] Graig Barter. D (2005) United State Pharmacopoeia (Indianapolis IN). **General Official Monography** for USP P. 27
- [4] Khashab M, Trector A. Z, and Kwo P.Y (2007) "Epidemiology of Acute Liver Failure" **Cur Gastroenterol Rep** **9**(1): Pp 66-73.
- [5] Lawrence K, & Beasley R (2008). "Association between Paracetamol use in Infancy and Childhood, rhino-conjunctivitis, and eczema in children aged 6 to 7 years" **The Lancet Journal** **37**(2):1039-1048.
- [6] Peter B. N. (1995) 'Drugs and Drug Action **Encyclopedia Britannica** 15thedition. Pp 529;530-560.
- [7] UKEssays. (November 2018). Paracetamol Synthesis Experiment. Retrieved from <https://www.ukessays.com/essays/chemistry/paracetamol> synthesis-experiment-2053.