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**Review Article**

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# Phosphoinositide 3-Kinase Family: Identification, Classification and Function in Human Therapeutic Issues: A Review

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**ABSTRACT**

*Phosphoinositide 3-kinases (PI3Ks), one of the families of eukaryotic enzymes was discovered more than 25 years ago and identified as a lipid kinase. PI3Ks can phosphorylate the 3-hydroxyl position of the inositol ring of phosphoinositides to generate lipid products and which involve in cell survival, growth, migration, apoptosis, proliferation, differentiation, intracellular trafficking and other cellular functions. In this review, the enzyme as a family comprising of 3 classes and 8 isoforms, the various domains found in the core structure of PI3K, the structural function of PI3K, its regulation and downstream targets were studied. It can be deduced that PI3K activity has a link with cancer and diabetes. It can be understood that PI3K pathway helps in the regulation of various cellular activities which leads to the protection of the immune system from malignant cells or pathogens, the pathway aids in the production of such malignant traits of the cancer cells and escape from immunity.*

**Keywords:** Phosphoinositide 3-kinases (PI3Ks); Phosphoinositides (PIs); Structure; Downstream target/receptor

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## INTRODUCTION

No cell lives on its own. In fact, cells respond to stimuli from the immediate environment. As seen in previous studies, cells in multicellular organisms survive, proliferate, differentiate or even die depending on intracellular and extracellular communication network. These cells have molecular switches that are formed from enzymes, regulatory proteins and receptors which aid in the amplification and integration of external signals [1]. Across the cytoplasm, it is interesting to know that lipid phosphorylation and hydrolysis plays a crucial role in the transmission of extracellular stimuli [2]. However, there is limited research on role of enzymes for these signalling events [2,3].

Lipids such as polyphosphoinositides constitute a small amount (~0.1%) of total cellular lipids and are involved in intracellular signalling [4].

One of the polyphosphoinositides known as Phosphoinosititde 3-kinases (PI3Ks) was discovered in 1984 with the identification of a minor inositol lipid kinase PI3K activity associated with polyoma middle T-Protein [5-9]. PI3Ks were also the premier mammalian lipid kinases discovered several years ago and presently, many more species of mammalian PI3Ks have been studied [10]. Whitman et al. [11] and Falasca et al. [12] also discovered that PI3K has the ability to phosphorylate the 3-hydroxyl position of the inositol ring of phosphoinositides [13]. As components of some signalling pathways, PI3Ks play vital roles in cell migration, growth, proliferation, differentiation, motility, survival, intracellular trafficking including phagosome formation [14] and autophagy [15], transport at the nuclear membrane [16], formation of internal vesicle within multivesicular endosomes [17] and other physiological events [6,8,18-22]. PI3Kinase activity has been linked with some human health problems such as cancer, diabetes, heart disease, allergy and inflammation. Many inhibitors of these lipid kinases have been employed for therapeutic purpose [12,20,23]. This review will focus on the overview of a family of lipid kinases called Phosphoinositide 3-kinases (PI3Ks) that are found in species spanning from murine to human and the therapeutic effects in human being. The family of PI3-Kinases will be classified based on its structure, function and disclose part of its regulation and downstream targets. By so doing, we hope to provide detailed information about PI3Kinase family.

## P13K

### Identification of PI3K

At first, PI3K had been identified as an 85-KDa (Kilo Daltons) phosphoprotein with its appearance correlated with the PI3K activity in immunoprecipitates especially from polyoma middle T mutants [24]. Domin et al. [2] reported the use of Polymerase chain reaction techniques and methods to isolate, amplify and characterize more novel PI3K family members by determining the sequence alignments with the kinase domains. Carpenter et al. [25] and Morgan et al. [26] studied and reported the formation of heterodimeric complex through cloning and purification of 110-KDa catalytic subunit (p110) and an 85-KDa adaptor (p85). There was also a sequence similarity between the cloned PI3K catalytic subunit and the catalytic domain in yeast protein, Vacuolar Protein Sorting 34 (Vps34p), which was involved in vesicular trafficking. On that note, the yeast also possessed PI3K activity [27].

More so, the cloning of the cDNA (complementary DNA) of Class II Human PI3K C2β containing C2 domain (conserved region) was achieved and the expressions of the enzyme (PI3K) in mammalian cells have been reported by Arcaro et al. [28]. Information generated and obtained has caused lack of understanding for those that are quite unfamiliar with this area of study. However, we will explain vividly about this family of lipid kinases and also use this knowledge acquired to proffer possible therapeutic solutions in further studies.

### PI3K Core Structure

Classification of PI3K family is mainly based on sequence alignments which defines and describes the four major homology regions (HR 1-4) of sequence similarity among members of class I family and other classes. As shown in Figure 1, HR1 (in grey colour) is the kinase core domain and it can be found in all PI3K classes. The kinase core domain displays weak homology to protein kinases [7]. HR2 (in green colour) is the Phosphoinositide 3 kinase, accessory domain (PIK domain) and it can be found in all four PI3K classes. This domain seems to act a scaffolding

role for the remaining domains [7]. HR3 (shown in small blue circle) is sometimes called the second conserved domain or protein structural domain (C2-like domain) and it can also be found in all the classes. In other proteins, C2 could mediate interactions with other proteins or lipids in a calcium dependent or calcium independent manner. The C2 domain is found in the PI3Kinase structure that suggests a role in substrate targeting and membrane binding [29]. Another C2 domain (shown in large blue circle) is present in class II PI3K only. HR4 (highlighted in red) is the Ras-binding domain which can found in Class I and II PI3Kinases only. Katso et al. [20] demonstrated that the HR4 had functional effects through interactions with Ras in Class I PI3Kinases. The x-ray crystals structures of the four domains (Ras, C2, Kinase core and PIK) from p110γ in Class $I_B$ PI3K have been determined and provides a proper understanding of PI3K at molecular level [29,30]. Other domains found in class $I_A$ PI3K are adaptor binding domain (ABD) that really binds to the regulatory units (p85 family members). The p85 units have five domains which include: Rho GTPase activating protein (Rho-GAP), N-terminal Src homology 3 (SH3), two SH2 (N-terminal nSH2 and the C-terminal cSH2) separated by an inter Src homology 2 (iSH2) domain that could bind to the catalytic subunit [31].

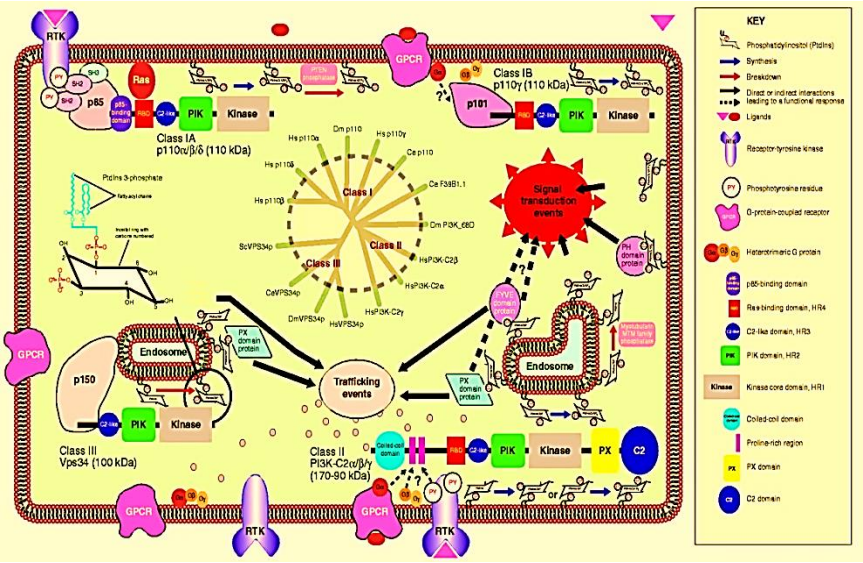



**Figure 1. Structure of Phosphoinositide 3-kinase family [34]**

**Classes of True PI3K**

This family of eukaryotic enzymes (PI3Ks) was grouped based on their substrate specificity and structural features [12]. According to Fry [6], about 12 members of PI3K family are present in the human genome and they can be divided into two main groups. They are the true PI3Ks and the PI3K-related enzymes. The true PI3Ks exhibit both protein kinase and lipid kinase activities and thus are divided into 3 classes which are Class I, Class II and Class III enzymes [13] whilst PI3K-related enzymes (Class IV enzymes) are proteins of a larger size that possess only the protein kinase activity [32]. The true PI3K family was grouped into three classes based on their basic structure; *in vitro* lipid substrate specificity, interactions with regulatory units and the additional protein domains present [33]. The three classes (I, II and III) of PI3Ks have eight isoforms [7] as shown in Figure 2. All classes of true PI3K possess C2 domain, catalytic kinase and helical domain [34] as shown in Figure 3. PI3Ks are partly based on their basic structures and substrate specificity. Thorpe et al. [35] reported that Class $I_A$ and $I_B$ PI3Ks phosphorylate

phosphatidylinositide 4,5-bisphosphate (PtdIns (4,5) $P_2$), and Class III PI3Ks phosphorylate PtdIns. They also contain some other domains like Phox homology (PX) domain, proline-rich (P) domain and C2 domains**.**

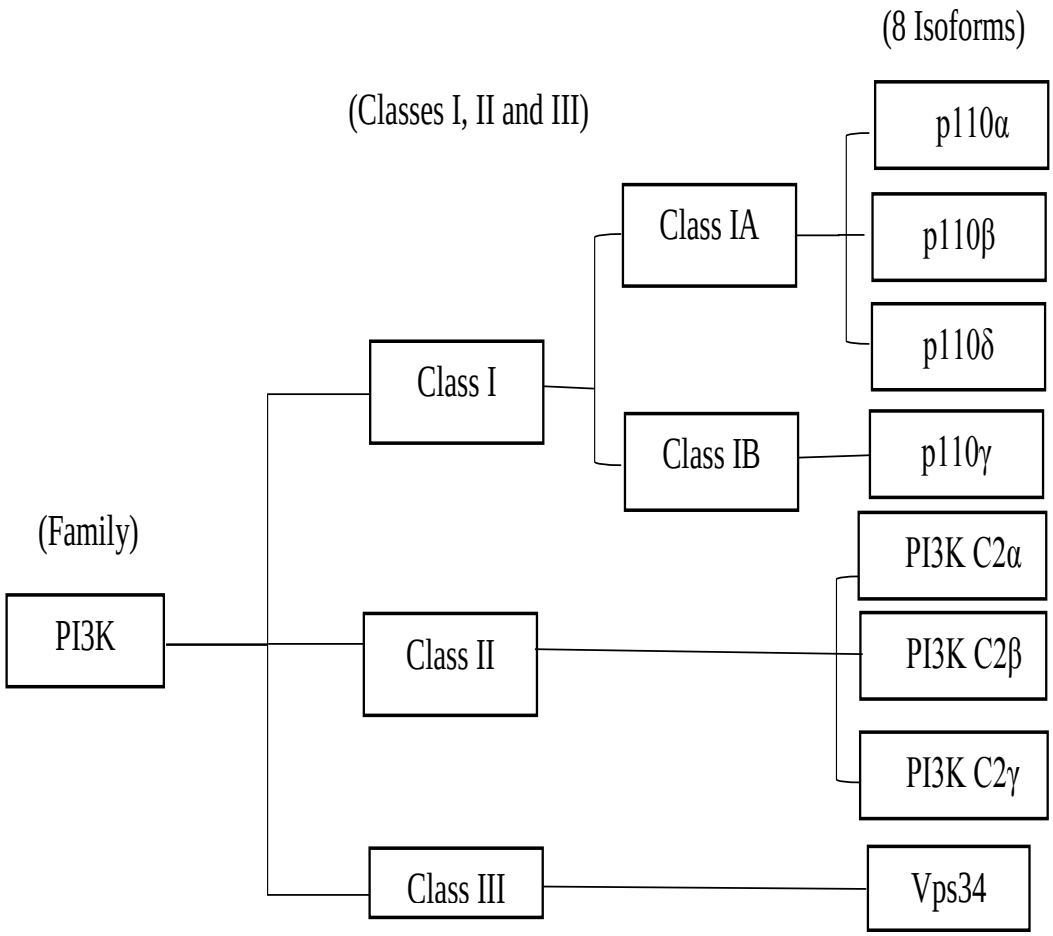



**Figure 2. The family, three classes and eight isoforms of this lipid kinase (PI3K)**

**Class I**

This class is the first class to be identified; the most studied and best understood class amongst all the classes which involved in human diseases such as cancers [36]. It was observed that a Class I PI3K was originally found associating with oncogene products and one member of this particular class was cloned first [18]. Class I PI3Ks are heterodimers consisting of catalytic subunit (110-120 KDa) and regulatory units (55-101 KDa). Many cell surface receptors can activate class I PI3Ks and some of them are G-protein coupled receptors, receptors with either associated or intrinsic protein-tyrosine kinase (PTK) activity or by action of small G-Proteins such as RAS [6]. Outside the cell, Class I phosphorylate Phosphatidylinositol [PtdIns], Phosphatidylinositol 4-monophosphate [PtdIns (4) P] and Phosphatidylinositol 4,5-bisphosphate [PtdIns (4,5) $P_2$] to generate Phosphatidylinositol 3-monophosphate [PtdIns (3) P], Phosphatidylinositol 3,4-bisphosphate [PtdIns (3,4) $P_2$ ] and Phosphatidylinositol 3,4,5-trisphosphate [PtdIns (3,4,5) $P_3$], respectively, but inside the cell, they phosphorylate PtdIns (4,5) $P_2$ to generate PtdIns (3,4,5) $P_3$ preferably, as shown in Figure 3 [6,37]. Class I is further divided into two subclasses which are Class $I_A$ and $I_B$ [38,39]. This division into two subclasses is based on the form of the adaptor subunit

associated with catalytic subunit [2]. This class has a link with the cell size in *Drosophila*. The p110α isoform can regulate the size of an adult heart in mammals [40].

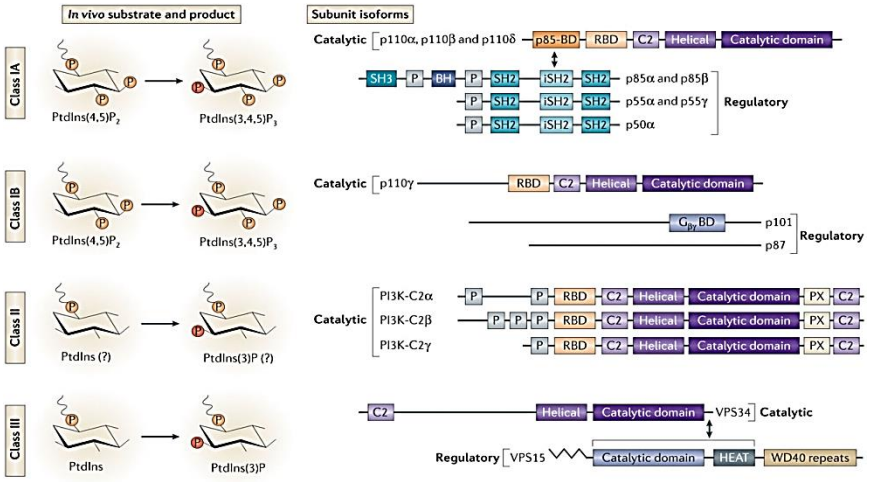



**Figure 3. A schematic representation of the three classes, subunit isoforms, in vivo substrates and products of PI3K [83]**

**Class $I_A$**

As rightfully said in 2.3.1, Class $I_A$ PI3Ks are heterodimers consisting of catalytic subunit (p110) and regulatory subunit (p50-p85). The catalytic isoforms are p110α, p110β, p110δ and the regulatory isoforms are p85α, p55α, p50α, p85β and p55γ as shown in Figure 2. Class $I_A$ PI3Ks have a Ras-binding domain, a helical domain, a catalytic domain and a p85-binding domain (p85-BD). Class $I_A$ PI3Ks possess an inter SH2 (iSH2) domain that separates the two Src homology 2 (SH2) domains [2] and usually binds to the catalytic subunits of Class $I_A$ PI3K [35]. The regulatory subunits consist of two Src homology 2 domains flanking a p110-binding site. The structurally conserved SH2 domain found in the Src oncoprotein exists in some intracellular signal transducing proteins. In human genome, the catalytic subunits are usually attached to the regulatory subunits. p85α (the most expressed regulatory subunit) and p85β were the first regulatory subunits to be identified [2] which contains Src homology 3 (SH3) and Src homology 2 (SH2) domains at N-terminal as well as breakpoint cluster homology (BH) domain in addition to other domains present as shown in Figure 2. The SH2 domains are about 100 amino acids in length whilst the SH3 domains are 60 amino acids in length. Individually, SH2 domain involves in communication between cells and SH3 domain interacts with peptide sequences rich in proline residues thereby acknowledging the existence of some cellular proteins [41]. Both Src homology 2 and 3 domains are involved in the regulation of Class IA PI3Ks by many activated protein tyrosine kinases (PTKs) [18]. Intrinsic ser/thr protein kinase activity found in Class $I_A$ PI3K phosphorylates the associated adaptor within $iSH_2$ of p85 especially in p110α and p110β [42] or gets involved by boosting auto phosphorylation in p110γ [43]. It has been reported that p110α plays a vital role in cell survival and p110β promotes cell proliferation [44]. Okkenhaug et al. [45] reported that_p110δ is responsible for B and T cell antigen receptor signaling.

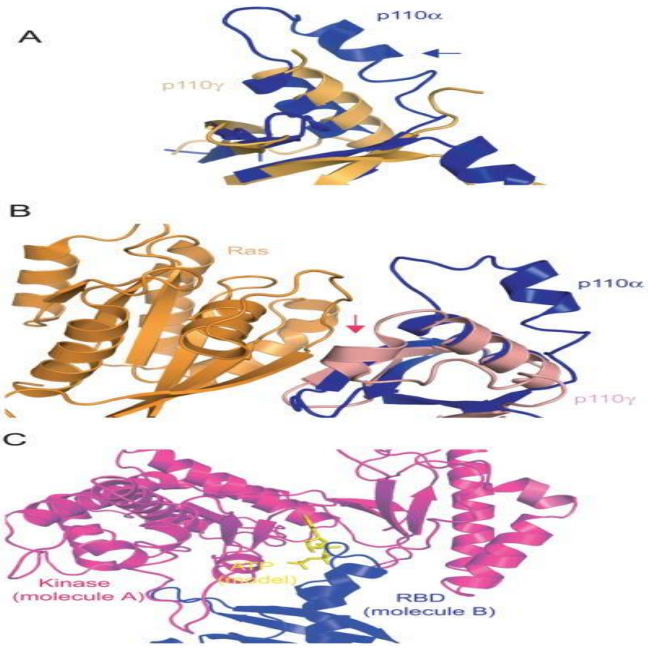
**Class $I_B$**

Class $I_B$ PI3Ks are also heterodimers consisting of a p110γ catalytic subunit and p101 or p87 regulatory subunit [46]. Class $I_B$ PI3Ks have a Ras-binding domain, a helical domain and a catalytic domain. The regulatory subunit, p101 contains no concluded or precise obvious signaling modules [6,7]. There is however, an indirect interaction between

Class $I_B$ PI3Ks and formyl-methionyl-leucyl-phenylalanine (FMLP), a type of G-protein coupled receptors [7]. In other words, G-protein-coupled receptor can activate Class $I_B$ PI3K through p101 regulatory unit [47]. A result from a research using knockout mouse models shows that p110γ acts as a great modulator of inflammation and allergy [48] and can also help to regulate cardiac contractility [40].

**Comparative Study of Class $I_A$ Pi3kα and Class $I_B$ Pi3kγ Crystal Structures**

Looking at the Ras binding domain (RBD) of p110γ, residues 255-267 are better arranged in the complex structure between p110γ and Ras rather than the free enzyme structure. On that note, it was proposed that Ras binding could result in ordering of this mobile loop but the residues 227-247 of p110α (corresponding subunit) is ordered in the absence of Ras and found to be in a conformation quite different from the one in p110γ-Ras binding structure as shown in Figure 4a and 4b. Also, some parts of RBD of p110α are attached and locked in the ATP-binding site of the kinase domain of a neighboring molecule in the p110α crystal structure as illustrated in Figure 4c. This interaction might represent a physiological interaction [31].




**A** = RBDs of p110α (in blue) + free p110γ (in gold)

**B** = RBDs of p110α + Ras/ p110 γ complex.

**C =** Kinase domain of p110α **(**molecule A, magenta) + RBD of p110α (molecule B, blue).

**Figure 4. (a-c) Comparison of the crystal structures of Ras-binding domains (RBDs) of p110α in Class IA PI3K and p110γ in Class IB PI3K [4]. The crystal structure (A) shows the comparison of free p110γ and RBDs of p110α. The blue arrow indicates the position of residues 227-247 in p110α. The second crystal structure (B) shows the relationship between the Ras/ p110γ complex and RBDs of p110α. The red arrow shows the arrangement of residues 255-267 in p110γ that become ordered by the binding of Ras. The last crystal structure (C) displays the interaction between the RBD and kinase domain of p110α. The RBD is attached to the ATP binding pocket of the kinase domain**

**Class II**

Class II PI3Ks are monomers with high molecular weight because of the extensions found at both N and C terminals [12]. Class II PI3Ks is the premier class to be identified by sequence homology amongst all the classes [12]. MacDougall et al. [49] discovered and identified the first isoform of Class II PI3Ks in an animal, Drosophila melanogaster. Class I and II are partly structurally related due to possession of particular class domains such as C2, helical, Ras binding proline-rich (P) and catalytic [6,34,46] but Class II PI3Ks lack p87 and p101 regulatory

subunits. Foster et al. [7] reported that C2 domains at the C-terminal present in other proteins act as a calcium-dependent phospholipid binding motif to mediate proteins translocation to membranes. As seen in Figure 1, the PX domains (in yellow) interact with PtdIns (3) *P* to target proteins to membranes [30]. Some basic functions of Class II PI3K are linked to receptor – mediated signalling processes such as epidermal growth factor (EGF) signalling, insulin, platelet - derived growth factor signalling and integrin signalling in platelet [6,50,51]. Class II PI3Ks are about 170-210KDa in size [7] and comprise of three isoforms. The isoforms of class two PI3Ks found in humans are PI3K C2α, PI3K C2β and PI3K C2γ [12,28,51-53]. Banigo et al. [13] successfully demonstrated and reported the qualitative PCR analysis of Human PI3K C2β C2 domain without additional magnesium ions. The successful purified will apparently be used for cloning in further studies. PI3K C2α and PI3K C2β can be found and grossly expressed in many mammalian tissues [52,54,55] whilst PI3K C2γ are present in a few tissues including the liver, prostate and breast [56]. It has been reported that Class II PI3Ks can phosphorylate PtdIns and PtdIns (4) *P* rather than PtdIns (4, 5) $P_2$ [6,7,57]. The determination of more basic functions and substrates (*in vivo*) of this particular class II PI3Ks is still in progress. Class II PI3Ks can be associated with intracellular membranes and plasma membrane [6] but may not necessarily be crucial for epidermal differentiation [58]. However, an epidermal growth factor which is an extracellular signal has been used in a way to stimulate a type of Class II PI3K (PI3K C2β) activity [59,60]. The first isoform of Class II PI3Ks, PI3K C2α, was identified as the target lipid kinase for a set of receptors in the plasma membrane and associated with clathrin-coated vesicles [61]. This isoform can also be resistant to wortmanin, an inhibitor of PI3K [50]. Investigation revealed that amongst all three classes of PI3K (Class I PI3K, Class II PI3K and Class III PI3K), Class II PI3Ks are the least commonly existing members.

**Class III**

Emr and colleagues identified the first member of this class as yeast protein, vacuolar protein-sorting 34 (Vps34) [12,27]. These enzymes phosphorylate PtdIns to generate PtdIns (3) *P in vitro* or *in vivo* only. Class III PI3Ks have catalytic, helical and C2 domains as shown in Figure 2. This class consists of just Vps34 catalytic subunit which makes use of PtdIns as a substrate and binds to Vps15 regulatory subunit [7,33]. Foster et al. [7] reported that Vps 15 contains a HEAT domain (suspected to mediate protein-protein interactions), a catalytic domain (which may be inactive) and WD 40 repeats (which possess structural and functional characteristics similar to a G-Beta subunit). The basic functions of Class III PI3Ks are autophagosome formation in human [36,62-64] and membrane trafficking regulation [33]. The only PI3K present in this yeast genome (*Saccharomyces cerevisiae* protein, Vps34p) is involved in trafficking and vesicle sorting [46,65]. It has been reported by Fry [6] that both human and yeast Vps34 proteins can be associated with the N-terminally myristoylated serine/threonine kinase (Vps 15/p150), which recruits the Vps34 to cell membranes and improves the activity of the lipid kinase'. Some of the Class III homologs reported in some eukaryotes include *Schizosaccharomyces pombe, Drosophila melanogaster* [66], plants [67], mammals [68,69] and *Candida albicans* [70].

**PI3Kinase Related Proteins**

These proteins are larger in size and are sometimes called Class IV PI3Ks. They are about 270KDa in size and possess a particular region homologous to the catalytic domain of PI3Kinase. Examples are DNA-dependent protein kinase (DNA-PK), ataxia telangiectesia-mutated gene product (ATM), ataxia telangiectesia-related (ATR) and the

targets of rapamycin (TOR) in mammals [71]. These proteins are serine/threonine protein kinase. ATM, DNA-PK and ATR can be involved in cell cycle checkpoint control, repair and maintenance of chromosomes (specifically when in contact with damaging agents such as ionizing radiation) while TOR proteins is involved in mitogenic and nutritional signals via the regulation of initiation for translation by the substrates p70 S6 kinase and sometimes the eukaryotic initiation 4E binding protein (4E-BP-1) and passage into the G1 phase of the cell cycle [6,32,72]. Most of Class IV enzymes have a few of the conserved motifs available in the catalytic domain of the true PI3Ks but do not have other domains such as PIK domain. Research have not shown that these proteins exhibit lipid kinase activity, but they possess protein kinase activity only as stated earlier in three classes of PI3Ks [6,32]. Fry [6] also reported that Class IV enzymes are quite sensitive to some PI3K inhibitors and could be found in various diseases such as cancer of different types and immunological diseases.

## THE REGULATION OF PI3KINASE ACTIVITY

In previous studies, Class I PI3Ks played vital roles in most receptor mediated signalling event [2] and binding of phosphopeptide to p85 SH2 domains generates an activation of lipid kinase activity [73]. Rodriguez-Viciana et al. [71] demonstrated that translocation to the plasma membrane draws the catalytic subunit close to its lipid substrate and also facilitates the interaction with GTP bound Ras, thereby, increasing PI3-kinase activity. This results to the pathway of class IA PI3-kinases parallel to the raf ser/thr kinase [74,75]. The study of the activation and regulation of PI3Kinase starting from class I molecules lead to a better understanding of class II and III PI3K through proper identification and development of appropriate reagents [2]. Regulation of P13kinase activity needs to be fulfilled for more insight on the physiological functions of PI3K class members.

## PI3KINASE DOWNSTREAM TARGETS

Domin et al. [2] reported that downstream targets could be called proteins as the identification of C2 domain and pleckstrin homology (PH) domain (lipid binding domains) support it. The suspected downstream targets of PI3K activity are as follow:-

### Rac

Rac is a member of Ras superfamily containing small GTPases and acts as a key downstream target of PI3K [2]. The three isoforms of Rac in mammals are Rac1, Rac2 and Rac3. Rac and Rho bind as well as activate (Phosphatidylinositol-4-Phosphate 5-Kinase) which gradually increases the amount of PtdIns (4,5) $P_2$. The Rac may also activate the JNK/stress activated protein kinase that phosphorylates c-jun most times [76]. c-Jun N-Terminal Kinases (JNKs) have been studied through isolation and characterization, as one of the very stress activated protein kinases that respond to various stress factors such as inhibition of protein glycosylation, heat shock, ultraviolet radiation and inflammatory cytokines when activated. Also, the neuronal growth factor (NGF) withdrawal-induced apoptosis of neurons occurred as c-Jun was activated. It was proposed that the inhibiting the function of c-Jun would eventually protect the neurons from the induced cell death [77]. JNK protein kinases are being encoded by three mammalian genes. JNK 1 and JNK 2 are usually expressed everywhere whilst JNK 3 can be found in the heart, brain and testis. The JNKs become activated when phosphorylation takes place on both threonine and tyrosine residues through MAP kinase kinases (MKK) 4 and MKK 7. MKKs can influence the regulation of the JNK signalling pathway which has been seen to be a complex one [77].

**p70S6 Kinase**

p70S6 kinase is a serine/threonine kinase which can be regulated by PI3K/mammalian targets of rapamycin (mTOR) pathway. This protein is another downstream target of PI3K. The target substrate required is the S6 ribosomal protein. p70S6 kinase plays a vital role in Gap phase1 to S phase transition in cell cycle and also phosphorylates the 40S ribosomal protein in response to mitogenic stimuli [78]. An important regulator of p70S6 kinase is the mammalian TOR (mTOR). The mTOR regulator does not directly phosphorylate p70Ss6. However, it is suspected that mTOR and PI3K activate p70S6 independently [2].

**Akt Kinase**

Akt Kinase also known as Protein Kinase B (PKB) is a serine/threonine kinase which acts as the cellular homologue of retroviral oncogene (v-akt) [79,80]. The Akt protein consists of protein kinase and N-terminal PH domain [81]. An example of Akt kinase, pyruvate dehydrogenase kinase 1 (PDK1), has been activated by both PtdIns (3,4) $P_2$ and PtdIns (3,4,5) $P_3$ and the subclasses of Akt are Akt1, Akt2 and Akt3. The GSK3 (an active protein kinase), acts as a major regulator in some physiological processes and whose activity is reduced by Akt. It acts as a negative inhibitor of c-jun and eIF-2B (transcription and translational factors) [82,83]. Akt activated by PI3Kinase will eventually lead to the inhibition and phosphorylation of GSK3 which results in the activation of some important regulatory proteins [2]. GSK has link to non-insulin-independent diabetes mellitus and production of neurofibrillary tangles (NFTs) could be associated with Alzheimer's disease [84]. Glycogen synthase kinase *3* (GSK-3) inhibitors can serve a vital role in the treatment of diseases caused by GSK-3 activity. GSK-3 inhibitors could be used and processed as potential drugs to treat Alzheimer's disease, diabetes, stroke and some other diseases [85].

**Protein Kinase C and Related Kinases**

Using *in vitro* method, the calcium independent isoforms of protein kinase C and other protein kinase C related kinases have been activated by 3' phosphoinositides [86,87]. A similar mechanism is suspected to be used in whole cells [2].

## PI3K AND BREAST CANCER

One major type of cancer considered in this review is breast cancer. Activation of PI3Ks can take place when there is signalling through the Epidermal Growth Factor (EGF) *family* of receptor tyrosine kinases (RTKs) [ErbB] family receptors especially with ErbB3, which usually have more than five (5) PI3K binding sites. Therefore, there is a possibility that in the presence of amplified ErbB family members in some breast tumours, enhanced PI3K signalling exist too [23,88,89]. Price et al. [90] and Ignatoski et al. [91] reported that enhanced migration of breast cancer cells has a link with the activation of PI3K either through overexpression of ErbB family receptors or growth factor activation. Fry [6] studied and reported that cytoplasmic *protein tyrosine kinase (*PTK), Proto-oncogene tyrosine-protein kinase Src, can be overexpressed in large quantity of breast cancers and via EGF, Src could sensitize cells to signalling. He also reported that breast tumour kinase (BRK), another cytoplasmic PTK, may also play an essential part in breast cancer. The overexpression of BRK could improve the joining of EGF signalling to PI3K/AKT by ErbB3 receptor phosphorylation. Three types of AKT proteins could be ubiquitously expressed in

some human tissues. AKT proteins can also be overexpressed in some human cancers including ovarian and breast. In all, overexpression or activation of PTKs and other proteins in breast cancer could actually lead to PI3K signalling in a large portion of breast tumours. Zhang et al. [92] studied and identified a signalling pathway in breast cancer namely PI3K/AKT/mTOR. This pathway plays a part in the inhibition of tumour cell apoptosis and tumour treatment, act as a new therapeutic target and a useful tool in breast cancer research. Zhang et al. [92] also confirmed that both mTOR and PI3K inhibitors could yield anticancer effect by considering targeting different levels of PI3K/AKT/mTOR signalling pathway during pre-clinical studies. Dituri et al. [93] reported that pharmacological inhibition of PI3Ks plays a vital role in diseases such as cancer because the tumours will stop growing and there will be a reduced immune-suppressive function mediated by PI3K. This could also cause more harm than good because PI3K signalling pathway is essential in antitumor immunity. More so, better and selective therapeutic inhibition of PI3Ks to target cancer cells without any negative inhibitory effect on the immune system should be considered.

## CONCLUSION

In conclusion, this study revealed interesting antioxidant and antimicrobial activities of the *Globularia alypum L* extracts in vitro assay. The present results showed that the ethyl acetate and butanol extracts manifested the strongest capacity to scavenge the stable DPPH free radical. More experiments in relation to this theme should be done to confirm the antioxidant activity of *Globularia alypum L*. Moreover all extracts remarkably inhibited the growth of all tested gram positive and gram-negative bacteria and proved to be an effective antifungal agent against tested fungi. In addition, we note that no previous studies on the antifungal activity of the plant extracts. These data confirm the great potential of this plant for the production of bioactive compounds, which can be suggested as a natural additive in food and pharmaceutical industries. However, further studies are needed to identify the compounds responsible for these beneficial properties.

## FUTURE RESEARCH

The determination of specific functions as well as potential binding partners of all the isoforms especially those in class II and III are of great importance. The fact that PI3K activity has a link with cancer and diabetes is a problem; so we must carry out more intensive research to study PI3K and the various types of cancer and diabetes involved and also proffer effective and efficient therapeutic procedures for that purpose. For example, the production of more inhibitors of PI3K and the determination of the efficiency of all of these inhibitors are of great importance. This review summarizes the identification and classification of PI3K also stated some functions of PI3K in solving human health issues.

## DECLARATIONS

**Ethics Approval and Consent to Participate**

Not applicable

**Consent for Publication**

Not applicable

**Competing Interests**

No Competing Interests

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