No: 657-Y
Please mark the appropriate section for this paper
☐ Experimental
☐ Clinical

CANCER GENOMICS & PROTEOMICS *22*: xxx-xxx (2025)
doi: 10.21873/cgp.xxxxx

Review

The WASP/WAVE Protein Family in Breast Cancer and Their Role in the Metastatic Cascade

RHIANNON YANNAN YU, WEN G. JIANG and TRACEY A. MARTIN

Cardiff-China Medical Research Collaborative, School of Medicine, Cardiff University, Cardiff, U.K.

Abstract

The Wiskott–Aldrich syndrome protein (WASP) and the WASP family verprolin-homologous protein (WAVE) family are essential molecules that connect GTPases to the actin cytoskeleton, thereby controlling actin polymerisation through the actin-related protein 2/3 complex. This control is crucial for forming actin-based membrane protrusions necessary for cell migration and invasion. The elevated expression of WASP/WAVE proteins in invasive breast cancer cells highlights their significant role in promoting cell motility and invasion. This review summarises the discovery, structural properties, and activation mechanisms of WASP/WAVE proteins, focuses on the contribution of the WASP/WAVE family to breast cancer invasion and migration, particularly synthesises the results of nearly a decade of research in this field since 2015. By exploring promising therapeutic strategies for breast cancer, including small molecule inhibitors and biological agents, this review stresses the potential for developing anticancer drugs that target the WASP/WAVE family and associated pathways, intending to improve the prognosis for patients with metastatic breast cancer.

Keywords: WASP, WAVE, N-WASP, breast cancer, migration, invasion, review.

Introduction

Breast cancer, the most commonly diagnosed cancer in women globally, results in a significant number of cancer-related deaths (1). This calls for advanced research, improved public health strategies, and enhanced treatment methods (2). Metastasis refers to the spread of cancer cells from the primary tumour to other parts of the body, affecting prognosis and complicating treatment (3). This review focuses on exploring the mechanisms underlying the metastasis process, with particular emphasis on the role of the actin cytoskeleton and

Dr. Tracey A. Martin, Division of Cancer and Genetics, School of Medicine, Heath Park, Cardiff, CF14 4XN, U.K. E-mail: Martinta1@cardiff.ac.uk

Received November 5, 2024 | Revised December 4, 2024 | Accepted December 18, 2024

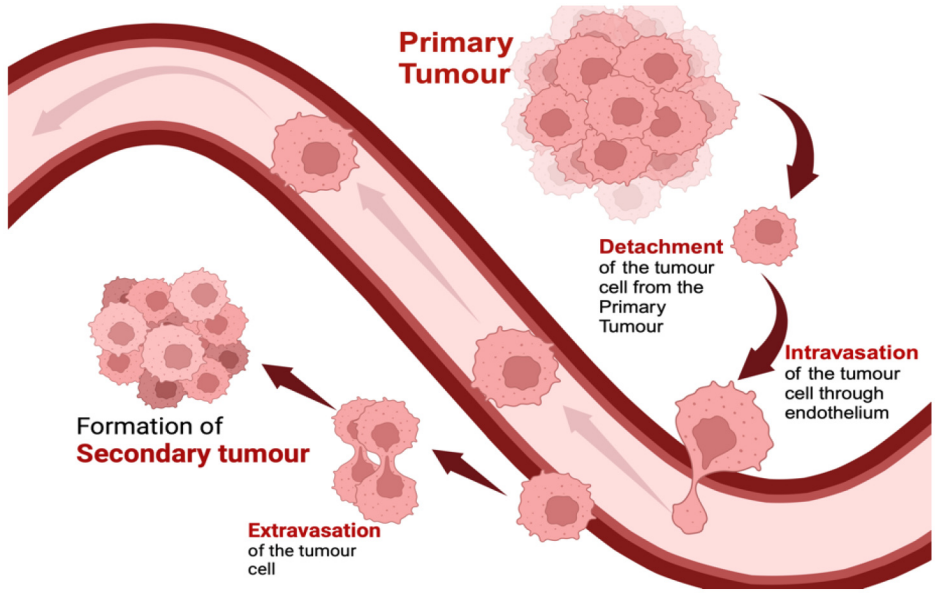

Figure 1. *The sequential steps of metastasis. Emphasizing the spread of cancer from its original location to new areas within the body. Cells from a primary tumour detach and invade through the endothelial lining of a blood vessel, a process known as intravasation. Once inside the bloodstream, these cells travel to distant sites. They then exit the bloodstream through extravasation. These cells then proliferate at a secondary site, forming a new tumour.*

WASP/WAVE family proteins in breast cancer invasion and migration.

Metastasis involves cell detachment, intravasation, extravasation and the formation of micrometastases (Figure 1) (4). These processes require the reorganization of certain key cytoskeletal components, particularly actin filaments whose dynamics are paramount in the remodelling of cytoplasm during increased motility characteristic of invasion (5). The proteins WASP/WAVE, central in the dynamics of actin and movement of cancer cells, act as molecular scaffolds to link migratory signals to activation of the Arp2/3 complex and enhance actin polymerization (6). This generates lamellipodia and invadopodia necessary for cell motility and degradation of ECM (7). Both WASP (comprising WASP and N-WASP) and WAVE (WAVE1, WAVE2, WAVE3) families are essential in breast cancer cell invasion and metastasis, coordinating signals for actin polymerization and protrusion formation aiding tumour cell invasion (8).

Studies (9, 10) indicate a higher expression of WASP/WAVE in invasive breast cancer cells, emphasizing their role in disease pathology. Down-regulation of these proteins could limit the required cytoskeletal changes for cancer cell migration, potentially leading to improved prognosis and survival rates in breast cancers (10, 11).

Understanding how these proteins regulate changes in the actin cytoskeleton in cancer cells is crucial for the development of specific therapies.

Methods

Literature search strategy. To systematically explore the roles of the WASP/WAVE family proteins in breast cancer, a comprehensive literature search was conducted using multiple biomedical databases, including PubMed, Web of Science, and Google Scholar. These platforms were chosen for their extensive coverage of molecular biology, cancer biology, and biomedical research.

To identify relevant studies, combinations of keywords and controlled vocabulary terms (*e.g.*, MeSH terms in PubMed) were applied. The search strategy included terms such as "Wiskott-Aldrich syndrome protein",

"WASP", "WAVE", "N-WASP", "actin cytoskeleton", "breast cancer", "metastasis", "cell motility", "tumour invasion", and "therapeutics". Boolean operators (AND, OR) were employed to optimise retrieval and refine results.

Inclusion and exclusion criteria. The following criteria were employed to ensure the selection of studies aligned with the objectives of this research:

Inclusion criteria.

1. Relevance: Studies investigating the structure, activation mechanisms, and functional roles of WASP/WAVE family proteins in breast cancer cell motility, invasion, or metastasis were included.
2. Study types: Original research articles and comprehensive reviews were prioritized, with a particular emphasis on studies using molecular or cellular models relevant to human breast cancer.
3. Language and accessibility: Only articles published in English and accessible through Cardiff University's institutional subscriptions were considered.
4. Recency and innovation: Preference was given to research published after 2010 to emphasize contemporary findings. However, landmark studies foundational to understanding WASP/WAVE functions were included where relevant.

Exclusion criteria.

1. Non-relevant topics: Studies unrelated to breast cancer or focusing on WASP/WAVE proteins in pathways unassociated with cancer metastasis were excluded.
2. Publication type: Non-peer-reviewed articles, such as conference abstracts or letters, were excluded unless they provided critical or emerging insights unavailable elsewhere.
3. Redundancy: Studies with overlapping findings were reviewed to extract unique contributions, but repetitive publications without novel information were excluded.

WASPs and WAVEs

Discovery and members of the family. In 1937, Alfred Wiskott first delineated the symptoms of microthrombosis, bloody diarrhoea, eczema, pyrexia, and recurrent ear infections in three brothers while sisters were not affected. He hypothesized it is a novel hereditary thrombophilia different from atopic thrombocytopenic purpura. Later, in 1954, Robert Aldrich studied a family where 16 out of 40 males over six generations showed similar symptoms and no female cases, demonstrating an X-linked pattern of inheritance. This condition was later named in honour of Wiskott and Aldrich as the great pioneers for its identification and description (12).

In 1994, Derry *et al.* (12) successfully isolated and identified the gene responsible for Wiskott-Aldrich Syndrome, hence naming it Wiskott-Aldrich Syndrome Protein (WASP), mainly found in the cytoplasm of hematopoietic cells (13). Two years later, a Japanese team found a 65-kDa brain protein, homologous to WASP but with a broader expression, particularly in nerve cells, and named it neural WASP (N-WASP) (14). Further research discovered a cDNA structurally homologous to WASP and N-WASP, resulting in the designation of the resultant protein as WAVE, for WASP-family verprolin-homologous protein, marking it as the third member of the WASP family (15). Further exhaustive database analyses identified two additional mRNAs encoding proteins related to WAVE, named WAVE2 and WAVE3, thus completing the five major members of the WASP family discovered hitherto.

This tremendous research has expanded the WASP family. New identifications in recent discoveries include junction-mediating and regulatory protein (JMY), Wiskott-Aldrich syndrome protein and scar homolog (WASH), and WASP homolog associated with actin, membranes, and microtubules (WHAMM). Also, among the recent discoveries are a couple of proteins, WASP and MIM-like (WAML) and WASP lacking the WH1 domain (WAWH). Due to the evolving nature and partially understood science of these new proteins, their mention will be brief in this regard (16, 17).

Structure

Members of the WASP family possess a common proline-rich domain (PRD) as well as a VCA region at the end of

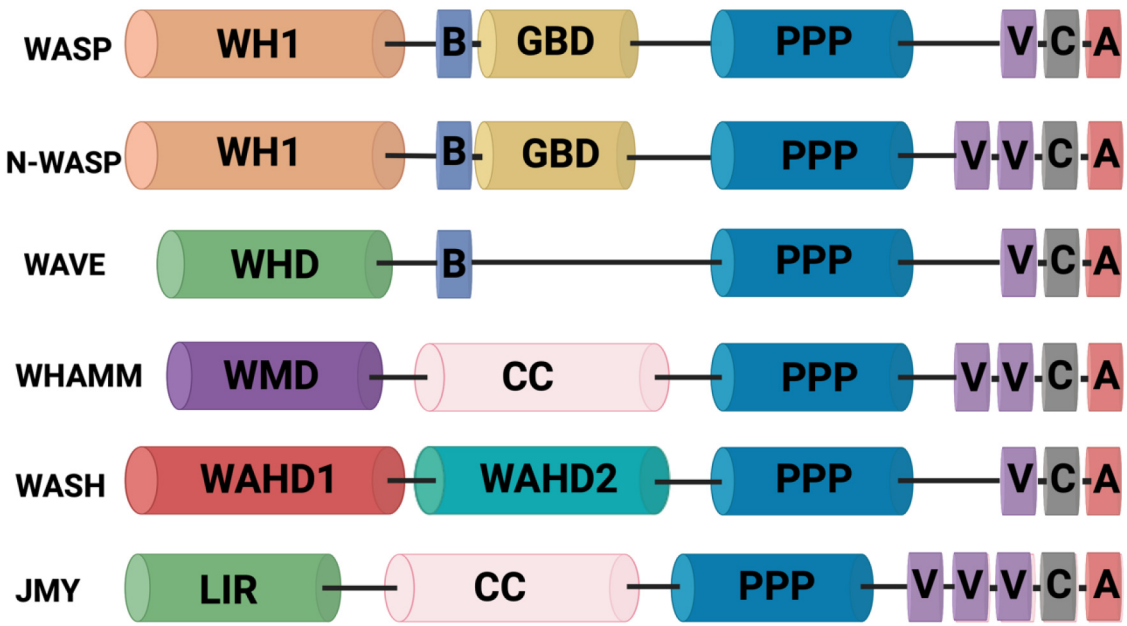

Figure 2. *The protein domains of individual members of the WASP family. The C terminus is highly conserved between the members, while the N terminus is divergent. WH1, WASP homology domain1; WHD, WAVE homology domain; WMD, WHAMM membrane interaction domain; WAHD1/2, WASH homology domain 1 and 2; LIR, LC3-interacting region; B, basic domain; GBD, GTPase binding domain; CC, coiled-coils domain; PPP, polyproline domain; V, verprolin homology domain; C, central domain; A, acidic domain. In general, the N-terminal domains are thought to confer spatiotemporal regulation on the protein, while the WCA regions at the C-terminal are required for activation of Arp2/3.*

their C-terminus. Each individual within this family exhibits a distinct N-terminus (Figure 2) (18). WASP and N-WASP exhibit substantial structural similarity, sharing 50% homology. Both possess identical functional domains and motifs, such as the EVH1 (or WH1) domain, a highly basic region, a GBD/CRIB motif, a proline-rich region, and a VCA region (19).

In mammalian cells, the EVH1/WH1 domain interacts with proteins from the WASP-interacting protein (WIP) family, including WIP, CR16, and WICH/WIRE (WIP- and CR16-homologous protein/WIP-related). WASP and N-WASP predominantly form stable complexes with WIP-family proteins, enhancing their stability, protection from degradation, and overall activity (20, 21). The basic region and GBD/CRIB motif of WASP/N-WASP facilitate activation by binding to PIP2 and active Cdc42, respectively. Numerous SH3 domain-containing proteins, such as Grb2/Ash, Nck, and WISH, interact with the proline-rich area, further promoting their activation (22). The C-terminal VCA region, comprising verprolin-homology (V), cofilin-homology (C), and an extremely acidic (A) area, is crucial for initiating Arp2/3-dependent actin polymerization *in vitro*. The V and A regions attach to actin monomers and Arp2/3, respectively. The C region interacts with Arp2/3, causing notable alterations in its tertiary and quaternary structures, thereby influencing its capacity to initiate actin polymerization (23).

Interestingly, N-WASP features two V-structural domains, in contrast to the single V-structural domain in WASP and WAVE. This distinction potentially explains N-WASP's enhanced efficacy in actin polymerization activation (24).

The WAVE protein, differing from WASP and N-WASP, lacks a GBD/CRIB motif. Instead, it harbours a WHC/SHC motif at its N-terminus, in addition to a fundamental domain, a domain rich in proline, and a domain VCA. Its C-

terminus shares significant similarities with proteins in the WASP family (25).

Activation of the WASP/WAVE Family

WASP and N-WASP exhibit autoinhibition, but WAVE is kept inactive by a combination of four additional proteins. WASP and N-WASP are folded through an interaction between the GBD/CRIB domain and the cofilin-homology (C) domain, which conceals the VCA region, and prevents the ARP2/3 complex from accessing the VCA region (8). Rac and Nck activate WAVE by disassembling the regulatory complex, so allowing WAVE to activate the Arp2/3 complex (26).

Where the C-terminal VCA domain binding to the Arp2/3 complex promotes its WASP activation and finishes the nucleation event, initiating actin polymerization and branched-actin networks assembly, the GBD domain in its N-terminal initially binds an activated Cdc42 or Rac1 GTPases converting it to an open conformation which presents the VCA domain This enables the V-domain to bind with the G-actin monomer, and the A-domain binds with the actin filament, and C-domain bind with the Arp2/3 complex within the VCA domain. Eventually, this promotes new actin filament nucleation through the conformational change carried out by the ARp2/3 complex (8, 27, 28).

Other WASP regions regulate the interaction of the VCA domain with the Arp2/3 complex. The BR domain interacts with negatively charged phospholipids at its N-terminus. The interaction includes phosphatidylinositol 4,5-bisphosphate (PIP2). PIP2 also activates WASP. The PPP domain binds to various signalling proteins, including SH3 domain-containing proteins, which recruit WASP to signalling complexes and regulate its activity (28). The activation of N-WASP is almost the same as that of WASP due to structural similarity (Figure 3) (29, 30).

WAVE proteins are regulated within a pentameric complex known as the WAVE regulatory complex (WRC) (31), it consists of WAVE itself, the specifically Rac1-associated protein 1/cytoplasmic FMR1 interacting protein 1 (Sra1/CYFIP1) subunit, Nck- associated protein 1 (Nap1) subunit, Abl-interacting factor (Abi) subunit, hematopoietic stem/progenitor cell protein 300 (HSPC300) subunit, and Nck-associated protein 1 subunit. In contrast to the activation mechanisms observed in WASP and N-WASP proteins, WAVE operates through a unique pathway significantly influenced by its interaction with Rac1, a small GTPase. The binding of Rac1 to WAVE triggers a series of conformational alterations within the WAVE regulatory complex (WRC), crucially leading to the unveiling of the verprolin-cofilin-acidic (VCA) domain. This exposure is critical for the activation of the Arp2/3 complex, thereby catalysing the initiation of actin polymerization (32, 33).

Central to this activation paradigm is the role of IRSp53, known as insulin receptor substrate p53, which fulfils a bifunctional purpose. Initially, it is instrumental in anchoring the WRC to the plasma membrane, a vital precondition for effective actin remodelling. Subsequently, IRSp53 serves as a molecular scaffold, enhancing the interaction between WAVE and various signalling entities, thereby fine-tuning WAVE's function (Figure 4). Among these regulatory interactions, the phosphorylation of WAVE by members of the p21-activated kinase (PAK) family stands out as a critical modulatory step (34). Such phosphorylation not only completes the activation of WAVE but also promotes the availability of the VCA domain. These intricate layers of interaction and modification underline the sophisticated regulation of WAVE, setting it apart from the activation routes of WASP and N-WASP. The nuanced control over WAVE's functionality is essential for the formation of lamellipodia and other actin-rich cellular structures, which are indispensable for cell motility and various cellular operations (35).

WASP/WAVE Protein Family in Cell Motility

The mobility of eukaryotic cells, a cornerstone for an array of critical physiological functions, is closely tied to the dynamic reconfiguration of the actin cytoskeleton (36). This framework of filamentous proteins is essential for sustaining cell shape, providing mechanical reinforcement, and facilitating motility (37). Its leading role is evident in

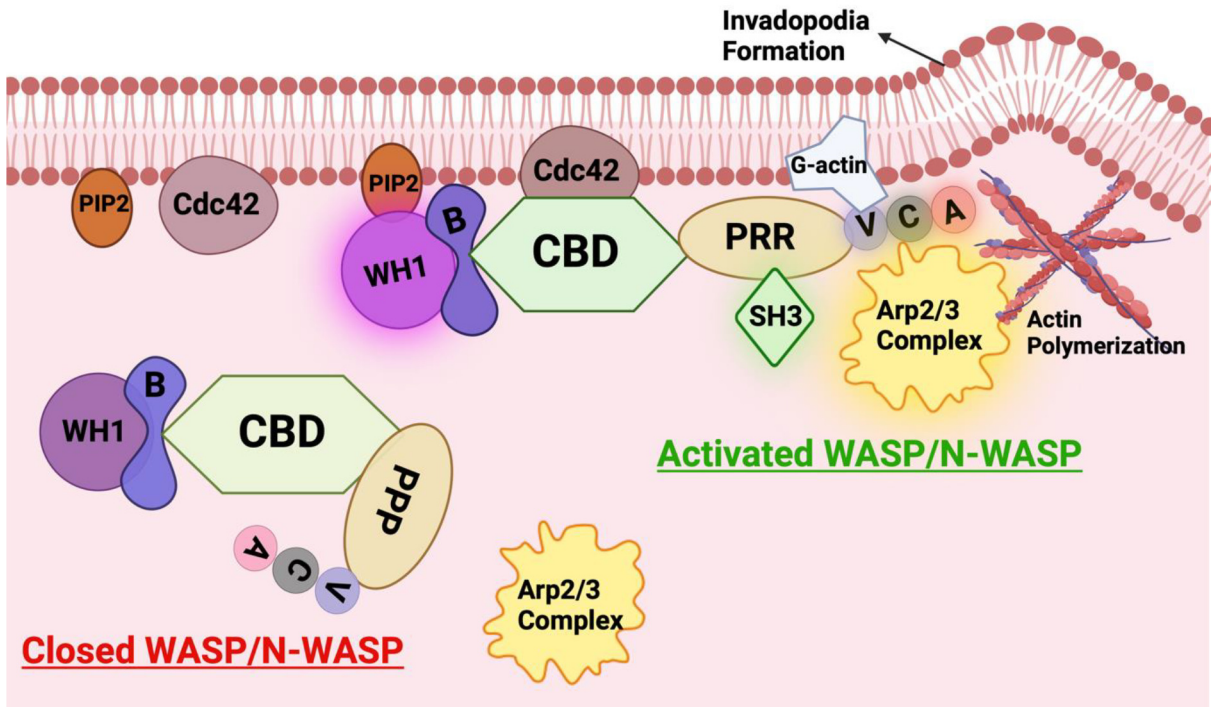

Figure 3. *WASP and N-WASP activation and their regulatory mechanism. (A) At resting state, WASP/N-WASP exist in an autoinhibited closed conformational state due to the interactions between members of the N-terminally located WASP homology domain (WH1) and the C-terminus. (B) the binding of Cdc42 to the GBD domain and the phosphoinositides (PIP2) to the WH1 domain exposes the VCA domain, the simultaneous binding of actin monomer (G-actin), and Arp2/3 complex activates Arp2/3 complex. The binding of the SH3 domain to the proline-rich region is important for Arp2/3 complex activation; (C) the active Arp2/3 complex leads to the activation of WASP/N-WASP; (D) the active WASP/N-WASP promotes promote the formation and elongation of actin filaments, contributing to actin polymerization.*

essential biological processes such as wound healing, immune system responses, and, most notably, in the progression of cancer metastasis, where the actin cytoskeleton's flexible nature enables cells to rapidly adapt to external cues and navigate complex environments effectively (38). Central to the organization of this cytoskeletal flexibility are the WASP and WAVE protein families. These entities play a key role in the generation of branched actin networks, interacting with the actin-related protein 2/3 (Arp2/3) complex, an interaction crucial for the development of lamellipodia and other cellular structures imperative for cell migration and the invasion of surrounding tissues (Figure 5) (26). Through facilitating these interactions, the WASP and WAVE families not only underpin cellular structure but also drive the sophisticated cellular behaviours necessary for navigating and adapting within the cellular landscape, showing their indispensable roles in modulating actin dynamics vital for a myriad of cellular processes and pathological conditions. In 2016, Weeks *et al.* (39) provided further insights into the functional roles of WAVE1 and WAVE3 in actin polymerisation and cancer metastasis. Using metastatic prostate cancer cells, they demonstrated that both WAVE isoforms interact with the ARP2/3 complex to regulate actin dynamics, contributing to the metastatic phenotype. Notably, WAVE3 knockdown significantly suppressed cell growth and enhanced tyrosine phosphorylation of ARP2, emphasising its regulatory role in ARP2/3 complex activity. Interestingly, suppression of cell invasion caused by WAVE1 knockdown was rescued by ARP2/3 inhibition,

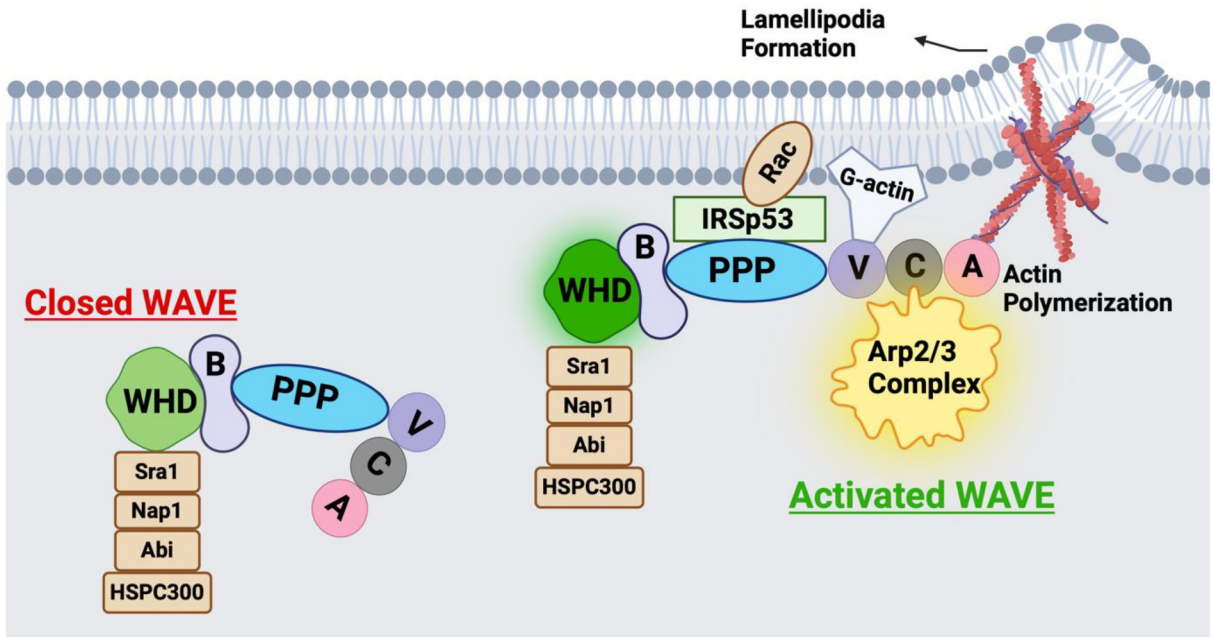

Figure 4. *WAVE activation and its regulatory mechanism. (A) At resting state, WAVE exist in an autoinhibited closed conformational state. (B) the N-termini containing the WHD domain that forms a protein complex by binding to Abi, HSPC300, Nap1and Sra1.The C-termini-containing VCA domain. IRSp53 recruits the WAVE complex to the plasma membrane. Upon Rac binding to the WAVE complex, the VCA domain of WAVE is exposed, making it binding and activating the Arp2/3 complex; (C) the active Arp2/3 complex activates WAVE; (D) the activated WAVE contributes to actin polymerization and leads to the formation of lamellipodia.*

highlighting functional distinctions between the two WAVE isoforms.

In environments such as the ECM or on culture dishes, where cells adhere and navigate in a bi-dimensional manner, the intricate mechanics of locomotion are predominantly reliant on the formation of lamellipodia and filopodia at the cell's leading edge (40, 41). Lamellipodia, characterized by their broad, leaf-like projections, are densely packed with branched actin filaments, a structure decisive for environmental sensing and propelling the cell forward (42). Conversely, filopodia are narrow, finger-like extensions enriched with tightly bundled actin filaments, acting as essential sensory tools. They excel in sensing and responding to chemical and physical cues from the ECM, thereby directing cellular migration and enhancing adhesion (43). The emergence of lamellipodia at the vanguard of migrating cells is fundamental to two-dimensional cell movement. Within these protrusions, actin branching, mediated by the Arp2/3 complex, forms a scaffold that advances the plasma membrane, propelling the cell (44, 45).

Although filopodia may not serve as the primary propulsion mechanism in cell motility, their role in mediating cell-environment interactions is critically important. Acting as cellular sensors, filopodia gauges external cues, and guides directional movement and adherence to the ECM (46). The rigid bundles of actin within these structures enable precise environmental sensing, highlighting their significance in complex processes such as tissue regeneration and the infiltration of cancer cells (47). The structural variation between lamellipodia and filopodia originates from their unique organization of actin filaments. Lamellipodia feature a dense network of actin filaments, initiated by the Arp2/3 complex, that supports the formation of a broad, advancing membrane front. This lattice is subject to continuous alteration, mirroring the cell's locomotive

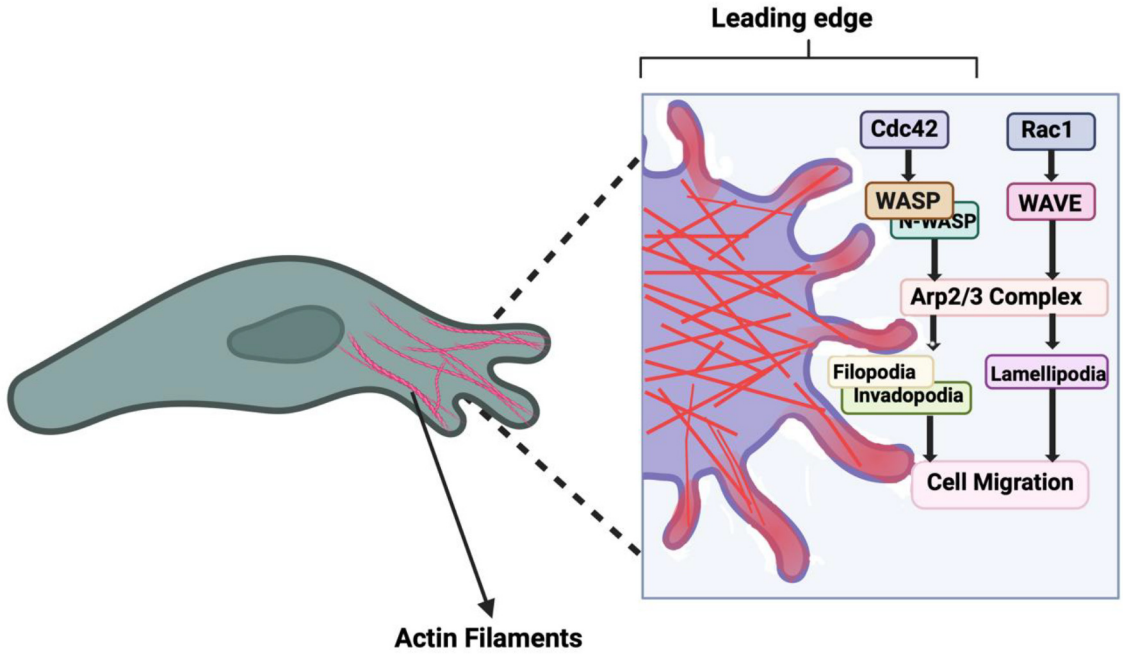

Figure 5. *Molecular factors involved in cell migration. Schematics illustrate the WASP protein family in actin filament organization at the leading edge of a cell, which is critical for cell migration. WASP and N-WASP, activated by the Rho GTPase Cdc42, facilitate the Arp2/3 complex's actin nucleation activity. Results in branched actin networks that propel cellular structures like filopodia and lamellipodia forward, driving cell movement. This process is vital for various cellular functions, including cell migration and wound healing.*

adjustments (48). In contrast, filopodia are formed by tight, aligned actin bundles, spurred by linear polymerization driven by formins and other actin-interacting proteins, providing the structural rigidity necessary for environmental exploration (49). The synergistic functions of lamellipodia and filopodia are essential for efficient navigation in planar spaces. Lamellipodia provide the necessary forward momentum, while filopodia guide the direction through environmental sensing (50).

The diverse functionalities of the WASP/WAVE protein family are intricately determined by the cellular environment and external stimuli, showcasing their involvement in a spectrum of cellular activities ranging from phagocytosis in immune cells to maintaining the morphology and structural integrity of epithelial cells (51, 52).

In three-dimensional matrices that emulate the complexity of *in vivo* environments, the significance of WASP/WAVE family proteins is magnified, particularly in cancer cell migration. These proteins are fundamental in arranging the formation of cellular extensions capable of navigating the ECM (53, 54). WASP/WAVE proteins also can construct actin-dense structures such as invadopodia, which are imperative for the degradation of and passage through the dense ECM. Through the regulation of actin polymerization and organization, these proteins not only facilitate the physical movement of cancer cells but also enable the cells to alter their immediate microenvironment, thereby promoting metastatic dissemination (Table I). WAVE2, in particular, is vital for lamellipodia formation across different cell types, with its deficiency significantly impairing these structures' development and, by extension, cell motility (8, 11).

Furthermore, the contribution of the WASP/WAVE family goes beyond merely aiding cell migration and invasion. These proteins are crucial for dynamic changes in cell shape and adherence, regulating the actin cytoskeleton. Through this regulation, WASP/WAVE proteins not only control the mechanical aspects of

Table I. *Functions of WASP protein family.*

Protein	Function	Role in cell motility
WASP	Induces actin polymerization for filopodia formation.	Senses environment, aids in cell-cell interaction and motility
N-WASP	Regulates actin dynamics, essential for invadopodia formation.	Promotes invasion through ECM, aids in metastasis
WAVE	Facilitates actin branching for lamellipodia formation.	Drives cell migration, invasion, and adhesion to ECM

cellular movement but also impact signalling pathways involved in gene expression, cellular survival, and proliferation (55, 56).

WASP/WAVE Protein Family in Cancer Cell Protrusions

Podosomes and invadopodia are specialized cellular structures decisive for the invasion and movement of cells through the extracellular matrix (ECM). These structures are key in breaking down the ECM, enabling invasive cells to navigate through tissue barriers. Found in a variety of cell types, including monocytic lineages, smooth muscle cells, and endothelial cells, podosomes act as multifunctional mediators for cellular navigation and engagement with the ECM. Characterized by their rich actin composition, these structures exhibit rapid cycles of assembly and disassembly, underscoring their role in cellular adhesion, signal transduction, and the exertion of mechanical forces (57). Such dynamic attributes are indispensable for the processes of cell migration and ECM interaction, as delineated in the research conducted by McNiven and Baldassarre (58). This suggests the critical function of podosomes and invadopodia in cellular dynamics and tissue invasion, stressing their importance in cellular motility and interaction with the surrounding matrix.

Invadopodia, structures uniquely characteristic of invasive cancer cells, bear both structural and functional resemblances to podosomes, yet are distinctly tailored to meet the aggressive demands of cancerous behaviour. These entities extend beyond mere cellular protrusions, serving as intricate biochemical platforms where various signalling pathways converge to govern the dynamics of actin. Research by V. Artym (59) and T. Meirson (60) has emphasised the proactive role of invadopodia in the degradation of ECM components, a central action in the progression of cancer metastasis. Invadopodia enable cancer cells to traverse the ECM, promoting invasive activities and the breach of tissue boundaries.

At the heart of invadopodia lies an actin filament core, essential for both its structural integrity and function. This core is encased by a multitude of adhesion proteins and proteases, indispensable for the dismantling of the ECM and thus aiding the invasive march of cancer cells (61). Moreover, the aggregation of diverse signalling molecules within invadopodia pointed to their significance not merely as physical extensions but as vital signalling centres (62). These molecules coordinate the response to both external environmental cues and internal cellular signals, tailoring invadopodia behaviour to navigate the complex and changing landscape of the cellular environment (63).

The genesis of invadopodia, important structures in invasive cancer cells, involves a sophisticated regulatory mechanism spearheaded by N-WASP and the actin-related protein 2/3 (Arp2/3) complex. N-WASP serves as a crucial mediator in this process, orchestrating the activity of the Arp2/3 complex, which is indispensable for the initiation and elongation of actin filaments. This interaction lays the groundwork for the development of the branched actin framework that is characteristic of invadopodia (64). In seminal research by Yamaguchi *et al.* (65) employing siRNA techniques, it was revealed that the attenuation of N-WASP expression significantly reduces the formation of invadopodia and adversely affects the capacity for ECM degradation, indicating the vital role of N-WASP in the mechanisms of cancerous invasion. Furthering this

understanding, Rotty and associates (52) provided compelling evidence of the synergistic function of N-WASP and the Arp2/3 complex in actin polymerization. Their research demonstrated that the interaction between N-WASP and the Arp2/3 complex is essential for generating the branched actin architecture, crucial for the structural integrity and functionality of invadopodia. This body of work collectively emphasizes the integral role of N-WASP in the dynamic assembly and operational efficacy of invadopodia, underlining its significance in oncogenic progression and metastasis.

WASP/WAVE Protein Family in Cancer Metastasis

The mechanisms of invasion manipulated by invadopodia and the N-WASP protein contrast with the roles of other family members like WAVE1 and WAVE2, which are deeply involved in controlling lamellipodia formation, as elucidated by Takenawa and Miki (66). This distinction within the WASP/WAVE family exemplifies the complex regulatory schemes that dictate actin cytoskeleton dynamics. Each protein's unique function mirrors a highly sophisticated regulatory network, ensuring specific cellular processes, ranging from migration to more aggressive invasion tactics, are finely tuned by the targeted activation and interaction of these proteins. Notably, the WAVE complex, comprising WAVE1 and WAVE2, collaborates with the Rac GTPase, playing a pivotal role in lamellipodia development (67). In a divergent manner, N-WASP leverages its interaction with the Cdc42 GTPase and the ensuing stimulation of the Arp2/3 complex to lead the charge in invadopodia formation. This distinction exhibits the varied roles these proteins play in cancer cell metastasis, demonstrating their specialized functions in modulating cell movement and invasion strategies (68).

Further investigations in this field, including a significant study by Oser (69), have contributed to our understanding of the role of N-WASP in cancer dynamics. By modulating N-WASP expression across various breast cancer cell lines, this research illuminated how a reduction in N-WASP activity significantly curtails both the formation and proteolytic capabilities of invadopodia.

Recent scholarly work further delineates the paramount importance of N-WASP and the Arp2/3 complex in the trajectory of cancer development. For instance, Lomakina (70) draws attention to the adverse prognostic implications in breast cancer patients associated with diminished levels of Arpin, a member of the inhibitory Arp2/3 complex family. Additionally, Kazazian (71) unveiled a novel function of the polo-like kinase 4 (Plk4) in modulating the Arp2/3 complex, thereby influencing cell motility in cancer scenarios. This discovery suggests Plk4's interaction with the Arp2/3 complex as a potential mechanism for altering actin cytoskeletal arrangements. In a similar vein, Laporte and Magistrato (72) employed all-atom simulations alongside dynamical network analysis to dissect the mechanisms governing Arp2/3 complex regulation. Their work lays a theoretical foundation for the development of small-molecule inhibitors aimed at curtailing cancer cell migration, presenting a novel strategy for intervention in cancer's invasive capacities.

Targeting the N-WASP pathway has implications beyond inhibiting cancer cell movement, it can disrupt dynamic interaction between cancer cells and ECM via inhibiting the formation of invadopodia, which could affect various stages of cancer progression, from tumour invasion to metastasis (73). Understanding and combatting cancer involves integrating research findings about podosomes, invadopodia, and the N-WASP pathway for a comprehensive view of cancer cell biology. Therefore, the N-WASP pathway is not just a molecular entity of academic interest but also a promising target for developing novel cancer therapies.

WASP in Breast Cancer

Up until now, detailed insights that are specific to WASP in breast cancer are somewhat limited. The focus tends to be broader, encompassing the entire WASP family proteins, which includes N-WASP and the WAVE proteins, due to their collective involvement in actin cytoskeleton

Table II. *Roles of WASP protein family in breast cancer.*

Protein	Role in breast cancer	Mechanisms and effects	Potential therapeutic implications
WASP	Modulates immune dynamics in the TME, influencing tumor progression and metastasis.	WASP drives macrophage chemotaxis within the TME, facilitating their accumulation at tumor sites where they can transition into a tumor-promoting phenotype known as tumor-associated macrophages (TAMs). These TAMs secrete factors that enhance tumor proliferation, angiogenesis, and the metastatic cascade.	Targeting WASP could disrupt pro-tumorigenic activities of TAMs, potentially reverting the TME to a more tumor-suppressive state. Precise targeting is required to minimize unintended immunosuppressive effects.
N-WASP	Plays a complex role, potentially acting as both a tumor suppressor and a promoter depending on the cellular context.	N-WASP is critical for the formation of invadopodia, crucial for ECM degradation and cell invasion. Overexpression or suppression affects the cell's ability to migrate and invade, impacting metastasis. nteraction with Claudin-5 affects cell motility and adhesion.	N-WASP may serve as a target for therapeutic interventions aimed at inhibiting cancer cell movement and metastasis, potentially through the modulation of its expression or activity.
WAVE	Facilitates cancer cell migration and invasion, particularly in aggressive forms such as TNBC.	Regulates the actin cytoskeleton to promote cell motility and invasion. High expression of WAVE proteins, especially WAVE2 and WAVE3, has been linked to poorer outcomes in breast cancer, indicating their role in tumor aggressiveness and metastatic potential.	WAVE proteins could be potential biomarkers for aggressiveness in breast cancer and targets for therapeutic intervention, especially in treatment resistant TNBC.

dynamics and cancer cell motility. However, the studies of Zicha *et al.* (74) and Ishihara *et al.* (75) collectively provide insight into the central roles of WASP in tumour metastasis, particularly emphasising the interplay between the immune system and cancer cells in breast cancer progression.

The tumour microenvironment (TME) constitutes a multifaceted ecosystem where cancerous cells, immune constituents, and stromal elements engage in dynamic interactions, profoundly impacting tumour evolution and the process of metastasis. Within this complex interplay, macrophages stand out due to their dualistic role that spans tumour suppression to the facilitation of tumour expansion and metastatic dissemination. Research by Zicha *et al.* (74) has displayed the critical function of WASP in driving macrophage chemotaxis (Table II). The study demonstrates that chemotaxis of macrophages is abolished in patients with Wiskott-Aldrich Syndrome (WAS), in these patients, while the speed of random motility of macrophages remains normal, their ability to move directionally in response to chemotactic signals is significantly impaired. This impairment is due to the disruption of the Cdc42-WASP-actin cytoskeleton pathway, The study's findings suggest that the lack of functional WASP leads to abnormal macrophage behaviour, which may contribute to the immune deficiency and eczema observed in WAS patients.

Ishihara *et al.* (75) further expanded comprehension of the intricate role that WASP plays in the immune system's contribution to breast cancer metastasis. They embarked on a study using mice that were selectively deficient in WASP within their hematopoietic cells, aiming to dissect the protein's impact on TAMs and their capacity to aid carcinoma cell invasion and spread. The absence of WASP in these specialized mice led to a marked reduction in the ability of TAMs to facilitate carcinoma cell invasion and metastasis, demonstrating the necessity of WASP for these processes.

To explore deeper into the underlying mechanisms, the researchers engaged in ex vivo studies that involved coculturing tumour cells with macrophages lacking WASP. This approach allowed them to directly observe the motility and invasion deficits in tumour cells that result from the absence of WASP in their macrophage counterparts. Their findings enlighten the key role of WASP in driving macrophage migration towards carcinoma cells that produce colony-stimulating factor 1 (CSF-1) and in the macrophage-mediated release of epidermal growth factor (EGF) through the action of metalloproteases. This release mechanism is integral for the promotion of breast tumour metastasis, emphasizing WASP's significant function in the intricate interplay between macrophages and carcinoma cells within the tumour microenvironment. This body of work not only elucidates a key pathway through which the immune system influences cancer progression but also identifies WASP as a critical facilitator of the metastatic process in breast cancer.

Beyond its role in immune cell function, WASP's involvement in actin cytoskeleton remodelling suggests direct implications for breast cancer cell motility and invasion. While Ishihara focuses on the leukocyte-dependent mechanisms of metastasis, the underlying principles of WASP-mediated actin dynamics are broadly applicable to cancer cell biology. It is plausible that WASP, by regulating actin polymerization and cellular protrusions, could directly influence breast cancer cell invasion and metastatic potential.

The dual role of WASP in both the immune response within the TME and direct cancer cell behaviour presents unique therapeutic opportunities. Targeting WASP could disrupt the pro-tumorigenic activities of TAMs, potentially reverting the TME to a more tumour-suppressive state. Furthermore, inhibiting WASP function in breast cancer cells could impair their motility and invasive capabilities, curtailing metastasis. However, given WASP's critical functions in normal immune responses and haemato-poiesis, therapeutic interventions would need to be precisely targeted to minimize unintended immuno-suppressive effects.

N-WASP in Breast Cancer

Martin *et al.* explored the impact of N-WASP in breast cancer (76). They suggested that N-WASP could act as a tumour progression inhibitor in human breast cancer (Table II). It was reported with lower relative expression of N-WASP in breast tumours versus the normal mammary tissues, and this has been associated with a reduced expression that is related to poor prognosis for the patients. Further critical support for the involvement of N-WASP in breast cancer was offered by *in vitro* experiments where it was demonstrated that invasive breast cancer cells with relatively higher expression of N-WASP showed reduced invasive and motile phenotype, implying its prospective clinical importance in breast cancer. Further research in breast cancer on the complex dynamics of Claudin-5, a tight junction (TJ) structure protein especially its interaction with the N-WASP signalling cascade was then done by the same team (77). In particular, they have portrayed a detailed relationship of Claudin-5 expression with balance in cell motility and adhesion using the MDA-MB-231 breast cancer cell line. Especially, the role of N-WASP during these regulatory processes was studied. Effects that were of particular interest in N-WASP inhibition in this study. Treatment of cells with a knockdown of Claudin-5 in MDA-MB-231 cells revealed that they blocked the response to N-WASP inhibition. This finding implies a direct relationship between Claudin-5 and N-WASP in signalling the motility of cells and hints at the potential role Claudin-5 may exhibit in human breast cancer metastasis through the N-WASP signalling pathway. This study evident that the N-WASP is a major regulator for the reorganization of the actin cytoskeleton and may turn out to be an attractive target for inhibition of cancer cell movement and subsequently inhibits cancer spread.

The role of N-WASP in promoting cancer invasion was further studied by Meirovitz *et al.*, who demonstrated that triiodothyronine (T3) activates the cortactin/N-WASP/ARP2/3 signalling pathway, facilitating actin cytoskeleton remodelling and focal adhesion formation in

breast cancer cells. These findings illustrate N-WASP's critical role in translating extracellular signals into cytoskeletal rearrangements (78).

Another research utilized MTLn3 rat mammary adenocarcinoma cell lines, manipulating N-WASP expression through over-expression of a dominant-negative construct and silencing via shRNA (79). These modifications resulted in a marked decrease in the cells' ability to form invadopodia and degrade ECM, demonstrating that decreased N-WASP activity impairs the cells' ability to migrate, invade, intravasate, and disseminate to the lungs. The study provides substantial evidence that N-WASP is crucial for the metastatic cascade in breast cancer, particularly in the early steps of metastasis, through its role in invadopodium formation and ECM remodelling.

Although these studies indicate differing perspectives on N-WASP's influence in regulating breast cancer cell movement, they concur on its potential association with breast cancer cell behaviour and the advancement of tumour development. The molecular mechanisms through which N-WASP operates, while still under investigation, have begun to reveal a complex network of interactions and activations. Of special relevance in this regard have been studies by Pichot *et al.* (68) proved N-WASP functions spanning from the activation of focal adhesion kinase (FAK) to the formation of invadopodia indispensable for the invasion of cancer cells through the extracellular matrix. N-WASP's activation and function are modulated by oestrogen receptor-α signalling and interaction with CIP4, an F-BAR protein that facilitates its localization to membrane sites crucial for cell movement.

The period following 2015 has witnessed critical developments in elucidating the function of N-WASP in the context of breast cancer. Extensive research has augmented our insight into the molecular basis of breast cancer progression, underscoring the potential of N-WASP as a therapeutic target.

A key focus has been the role of N-WASP in promoting cancer cell invasion and metastasis. Hebbrecht *et al.* (80) focused on the C-terminal VCA domain of N-WASP, demonstrating how VCA nanobodies can inhibit invadopodium formation by disrupting N-WASP's interaction with the Arp2/3 complex, thereby reducing matrix degradation activities of cancer cells. The recent study by Chung *et al.* (81) pointed up the emerging antiproliferative role of N-WASP in cancer biology. For squamous cell carcinoma (SCC), N-WASP particularly shows a significantly inhibitive effect concerning to proliferation and metastasis of carcinoma cells. This attenuation is mediated via an ERK2-dependent pathway, where N-WASP augments ERK2's phosphorylation of FOXO1, subsequently leading to an increase in TXNIP expression. These interactions suggest a complex regulatory mechanism where N-WASP functions as a potential tumour suppressor.

Another critical area of research has been the relationship between N-WASP and cancer cell resistance. Recently, the role of N-WASP in breast cancer resistance to natural killer (NK) cell-mediated cytotoxicity has been increasingly recognized. N-WASP facilitates the formation of an 'actin response' at the immunologic synapse between breast cancer cells and NK cells. This actin response is characterized by rapid F-actin accumulation, leading to a significant reduction in the cytotoxic protease granzyme B within the target cells and thereby decreasing apoptosis rates. Knockdown of N-WASP disrupts this actin remodelling, substantially enhancing the susceptibility of previously resistant cancer cells to NK-cell-mediated lysis. The study by Al Absi *et al.* (82) provides compelling evidence of this mechanism, demonstrating N-WASP's critical role in the modulation of immune evasion pathways in breast cancer.

The hormonal regulation of N-WASP has emerged as a significant area of interest. Mondaca *et al.* (83) highlight and discuss the molecular actions bestowed by those hormones – mainly luteinizing hormone (LH) and oestrogen – which contribute to breast cancer cell motility and invasion. These hormones in their turn generate complex pathways of signalling via their receptors bound together by the rearrangement process carried out with the actin cytoskeleton at different moments. Cortactin and N-WASP tend to be major players in both pathways during these

processes. LH, as demonstrated in a study on T-47D human breast carcinoma cells, initiates a signalling cascade involving kinases like Src and FAK, leading to the activation of cortactin and the Arp2/3 complex, thus facilitating cell migration and invasion. Similarly, oestrogen's effect, as shown in MCF-7 human breast cancer cells, entails the phosphorylation of paxillin, a protein essential for focal adhesion, which in turn regulates the N-WASP-Arp2/3 axis, modulating cytoskeletal dynamics and consequently influencing cell adhesion, migration, and invasion (84).

Decades of continuous research on Neural Wiskott-Aldrich Syndrome Protein have illustrated its potential as a therapeutic target in breast cancer, which has been increasingly recognized. N-WASP inhibitors, such as Wiskostatin, have been shown to affect various aspects of cancer progression including cell motility, invasion, and metastasis (85).

WAVE2 in Breast Cancer

Fernando *et al.* (86) initiated this exploration by demonstrating the elevated expression of WAVE2 in breast cancer tissues compared to normal breast tissues. Their study, using quantitative PCR and immunohistochemistry, linked high WAVE2 expression to poorer outcomes in breast cancer, specifically in node-positive cases and more advanced tumour grades. This landmark research proposed the possibility of WAVE2 as a prognostic marker for breast cancer and set grounds to launch future inquiries for its role in cancer biology. Continuing similarly, Koike and colleagues (87) clarify the predictive significance of cellular dissociation in cancer recurrence among individuals diagnosed with breast cancer. Their findings indicate that cellular dissociation, especially when associated with co-expression of Arp2 and WAVE2, is an independent significant predictor of recurrent disease. This study also emphasizes the importance of cellular morphology and molecular markers in understanding breast cancer behaviour and potential recurrence.

Expanding on the intricate dynamics of breast cancer cell behaviour, a study into the effects of pterostilbene on associated cellular mechanisms revealed the compound's potent inhibitory action on the Rac1/WAVE2/Arp2/3 signalling axis (88). This pathway's blockade holds considerable significance, given that the activation of the Rac1/WAVE2/Arp2/3 cascade is pivotal for the genesis of new actin filaments. The role of γ-tocotrienol on the Rac1/WAVE2 signalling pathway in mammary cancer cells detailed in another study, further expands on the theme of natural compounds in cancer treatment (89). The impact of rhapontigenin on breast cancer cells is similarly notable (90). Rhapontigenin effectively suppresses cell migration and invasion by inhibiting the PI3K-dependent Rac1 signalling pathway. This study contributes to the increasing comprehension of the impact that natural compounds can have on key signalling pathways within cancerous cells. In a study exploring the WAVE2/miR-29/Integrin-β1 oncogenic signalling axis, a new dimension of WAVE2's role in breast cancer, particularly TNBC, is unveiled (91). Additionally, research on the complex formation between WAVE2 and PKA in breast cancer cells provides a deeper understanding of WAVE2's involvement in cancer metastasis. This study shows that WAVE2 can act as an A-kinase anchoring protein (AKAP), recruiting PKA to specific subcellular locations, which influences the formation and enlargement of membrane protrusions essential for cell migration and invasion (92).

Very recently, Rana *et al.* (93) outlined the multifaceted role of WAVE2 in cancer biology by detailed review, which includes its profound effect on TNBC – well characterized for invasiveness and non-responsiveness toward currently available molecular therapies. Delving deeper into the molecular pathways regulating WAVE2, such as protein kinase B (AKT), TGF-β, VEGF, and ERK-MAPK pathways, which emphasized that the targeting of WAVE2 may be of high therapeutic potential in dealing with aggressive types of cancers.

WAVE3 in Breast Cancer

Sossey-Alaoui has been investigating the role of WAVE3 in TNBC progression and metastasis for the last 20 years, and

a series of insights in this area have been published. His 2005 study brings substantial progress in understanding the role of WAVE3 in cancer metastasis (94). He investigates the complexities of WAVE3 function in cell motility and invasion, in particular of MDA-MB-231 breast cancer cells. The study employed RNA interference to knock down WAVE3 expression and observed impaired expression levels of MMP-1, MMP-3, and MMP-9, indicating a crucial role of WAVE3 in regulating these MMPs, which are key promotors in cancer cell invasion and metastasis. As a consequence, cell motility and invasion were inhibited, coupled with increased actin stress fibre formation and reorganization of focal adhesion complexes. Crucially, the suppression of WAVE3 decreased phosphor-p38 MAPK levels without affecting the levels of phospho-AKT, phospho-ERK, or phosphor-JNK. This suggests that WAVE3 influences cell motility and invasion through a pathway involving p38 MAPK but not the other MAP kinases or AKT. Following this pioneering research, his 2007 study provides an in-depth investigation into the role of WAVE3 in breast cancer progression and metastasis and reports that down-regulation of WAVE3 would significantly affect tumour growth, invasion, and metastasis (95). There has been a considerable decrease in the invasive capacities of MDA-MB-231 cells following the suppression of WAVE3. Furthermore, a noteworthy decline in the growth and spread of tumours in animal models has also been demonstrated. The study also correlates high WAVE3 expression to be related to advanced breast cancer stages, thereby indicating it might be useful in the identification of tumour progression. The same year, he demonstrated that STI-571 (Imatinib Mesylate) effectively blocks the phosphorylation of WAVE3, underscoring the essential roles of both the WAVE3-Abl kinase interaction and Abl's kinase function in this process. This revelation identifies a precise molecular route in which Abl kinase activation of WAVE3 would enhance actin cytoskeleton dynamics, subsequently promoting cancer cell motility and invasiveness. Given STI-571's targeted inhibition of Abl kinase, employing specific inhibitors that disrupt the WAVE3-Abl connection or directly curb Abl's enzymatic action presents a promising strategy to thwart cancer progression (96).

Intrigued by those previous findings, in 2009, he employed quantitative RT-PCR to measure miRNA and mRNA levels, alongside Western blot analysis for protein expression and utilized luciferase reporter assays to verify the direct targeting of WAVE3 by miR200. Additionally, the impact of miR200 on cancer cell invasiveness was rigorously evaluated through cell invasion assays. The findings reveal a notable inverse relationship between miR200 expression and WAVE3 levels across various cancer cell lines, provides a comprehensive analysis of the regulatory dynamics between miR200 microRNAs and WAVE3, showcasing that miR200 directly targets and diminishes WAVE3 expression, thereby influencing cancer cell invasion and altering cell morphology (97).

Continuing on this exploration of how microRNA would influence the expression of WAVE3, the 2011 study provides a comprehensive analysis of the regulatory dynamics between microRNA miR-31 and the WAVE3 protein, in cancer invasion and metastasis (98). This work establishes a clear inverse correlation between miR-31 and WAVE3 expressions in both invasive and non-invasive breast cancer cell lines. MiR-31 directly targets WAVE3's 3'-UTR, suppressing its expression and thereby significantly diminishing the invasive phenotype of cancer cells. This interaction not only, delineates the regulatory mechanism of WAVE3 by miR-31, positioning WAVE3 downstream of miR-31, but also emphasizes the therapeutic potential of targeting the miR-31-WAVE3 axis to mitigate cancer cell invasion and metastasis.

The comprehensive review culminating in 2012, offers a comprehensive review of WAVE3's role in cancer progression and metastasis. It synthesizes a decade's worth of research on WAVE3, exploring its regulation in both normal physiological and pathological conditions (99). The review details WAVE3's involvement in actin cytoskeleton remodelling, interactions with microRNAs, and impact on cancer cell invasion. The potential application of WAVE3 as both a therapeutic target and a biomarker in breast cancer is also examined in the discussion, emphasizing its significance in subtypes of breast cancer that are characterized by aggressiveness.

Several other researchers from across the globe too have thrown light on the basic role of WAVE3 in breast cancer. Kulkarni *et al.* (100) have established a connection between higher levels of expression of WAVE3 and the progression and spread of TNBC. WAVE3 has been denoted as a predictive marker for cancer-specific mortality and metastatic potential, more prominently in TNBC. Taylor *et al.* (101) further investigate the correlation between TGF-β and WAVE3 in TNBC. They identify that TGF-β up-regulates WAVE3, associating it with the metastatic potential of TNBC cells and underscoring its role in EMT and metastasis. Both of the studies convincingly proved that WAVE3 not only contributed to the aggressive behaviour of TNBC cells but also the elevation of WAVE3 levels correlated with poor prognosis and treatment outcome. Therefore, it seems to be of utility as a therapeutic target in TNBC. Spence and colleagues (102) proceeded to advance this investigation, in which they further explored the involvement of Scar/WAVE3 in the mobility of breast cancer cells. Their research revealed that while Scar/WAVE3 is crucial for dynamic lamellipodial activities and cell motility on two-dimensional surfaces – manifested by the formation of larger, more stable lamellipodia and a reduction in cell speed – it does not significantly influence the invasive capabilities of cancer cells in three-dimensional matrices, marks the differential roles of Scar/WAVE3 in cell movement across varied environments.

Particularly, Wang *et al.* (103) specifically investigated the significance of WAVE3 phosphorylation in facilitating the coordination among the PI3K, TGF-β, and EGF signalling pathways. His study has indicated that WAVE3 phosphorylation is required for its oncogenic activity. It is established that WAVE3 can be phosphorylated downstream of PI3K, TGF-β, and EGF signalling pathways. This phosphorylation is not only the downstream event of these pathways but also importantly helps in its activation leading to the positive feedback loop further augmenting the progress of cancer. In vitro studies of the research showed that loss of WAVE3 phosphorylation greatly reduces migration of cells, growth of the tumorsphere, and invasion, thus showing the requirement of this post-translational modification for the behaviour of cancer cells. In vivo experiments using mouse models of breast cancer further substantiate the role of WAVE3 phosphorylation. Studies have reported that neoplasms originating from breast cancer cell lines exhibiting phosphorylatable WAVE3 (W3-WT) displayed heightened levels of phospho-AKT and phospho-SMAD in comparison to their non-phosphorylatable counterpart (W3-Y4). Consequently, this infers that the phosphorylation of WAVE3 constitutes a critical occurrence necessary for the initiation of downstream effector signalling. Only very recently, Kansakar *et al.* (104) probed WAVE3 phosphorylation within the proline-rich domain (PRD). They punctuated it as very important for malignancy-linked activities of WAVE3 in breast cancer, such as migration, invasion, and tumour growth and metastasis.

In 2020, a review summarizes recent advances in understanding the molecular mechanisms by which WAVE3 contributes to cancer progression (105). It discusses the regulation of WAVE3 expression and function, and the signalling pathways involved. The transforming growth factor-beta is one of the many multifunctional cytokines that determine cellular functions. In the context of cancer, this TGF-β signalling takes up a double role, that of a tumour suppressor role in the early stages, whereas at later stages of tumorigenesis promoting metastasis. Note, it has a vital role in up-regulate the expression of WAVE3 by activation of the p38 MAPK pathway which is an important signalling pathway as far as cell differentiation, apoptosis, and autophagy are concerned. This activation leads to the increased transcription of WAVE3, elevating the metastatic capability of cancer cells. The role of WAVE3 as a facilitator of TGF-β-driven metastatic activity in cancer accents its potential as a target for therapeutic intervention to curb cancer spread and progression. Further, the review outlines the interaction of WAVE3 with the Abl non-receptor tyrosine kinase that is essential in coordinating cell movement and the positioning of focal adhesions and lamellipodia. Specifically, Abl kinase directly phosphorylates WAVE3, activating it to regulate actin polymerization, which

in turn, influences the formation of lamellipodia and the dynamic assembly of focal adhesions. These focal adhesions serve as crucial points of contact between the cell and its external environment, facilitating signal transduction that informs cell direction and velocity. The precise modulation of these structures is integral to cell motility, impacting processes such as wound healing, immune cell migration, and cancer cell invasion.

WAVE3 was also identified as an important regulator in the progression of TNBC and treatment resistance. Although chronic exposure to chemotherapeutic agents does not lead to up-regulation of WAVE3 in breast cancer, WAVE3 expression is associated with chemoresistance through mechanisms involving the STAT1-HIF-1α-VEGF-A signalling axis (106). The study by Wang (107) illustrates the role of WAVE3 in stabilizing WAVE3-mediated stabilization of β-catenin leads to activation of the pathways of oncogenic signalling, that result in cell survival, and chemoresistance. WAVE3 likewise impacts the YB1-dependent activity of the transcription factor that triggers genes specifically expressed in cancer stem cell-like cell populations. The YB1 regulates the expression of β-catenin, thereby influencing the invasive capacity of triple-negative breast cancer (TNBC) tumours. Their findings further implicate that dual blocking of WAVE3 expression or phosphorylation, besides chemotherapy, will work in the direction of down-regulation of activity as well as expression of β-catenin, ultimately causing the inhibition of oncogenic behaviour of the chemoresistant TNBC cells. Moreover, recent findings by Mustafa Qasim *et al.* (108) revealed that reduced KISS1 expression – a known suppressor of metastasis – correlates with increased WASF3 (WAVE3) activity, suggesting a regulatory axis that promotes invasive phenotypes in TNBC cells. This was further supported by findings where the loss of KISS1 leads to up-regulation of ZEB1/2 and increased MMP9 activity, hallmarks of epithelial-to-mesenchymal transition (EMT) and extracellular matrix degradation. This study emphasizes the need to explore therapeutic strategies targeting the KISS1-WAVE3 signalling axis as a potential approach to mitigate TNBC progression.

Conclusion

This comprehensive review has explored the intricate roles played by the WASP/WAVE family of proteins within breast cancer invasion and metastasis. Central to the discussions are the mechanisms by which these proteins influence the actin cytoskeleton's dynamic reorganization, thereby facilitating the motility of cancer cells. The elevated presence of WASP/WAVE proteins in invasive breast cancer cells brings out their significant contribution to the disease's pathology. Through detailed discussion, this paper has illustrated the structural and functional nuances of these proteins, reveals their activation mechanisms and the consequential modulation of actin dynamics. Furthermore, it has broadened the scope of their impact, extending beyond the realm of cancer cell mobility to encompass roles in immune response modulation and maintenance of epithelial cell integrity.

Notwithstanding the depth of investigation, the research of the WASP/WAVE family acknowledges several inherent limitations. Primarily, the emphasis on WASP/WAVE proteins potentially obscures the roles of other cytoskeletal regulators in metastasis. Moreover, the transformation from research findings to clinical application remains embryonic, with a substantial need for *in vivo* studies and clinical trials to substantiate the therapeutic targeting of these proteins.

In the path toward clinical translation, the inhibition of WASP/WAVE activity stands out. Considering their essential function in the reorganization of the actin cytoskeleton and subsequent cell motility, the development of small molecule inhibitors or biological agents that can effectively modulate the activity of WASP/WAVE proteins represents a viable strategy to curb cancer cell migration and metastasis. Alternatively, targeting the regulatory pathways that control WASP/WAVE functions could indirectly hinder their activity. For instance, impeding the interactions between these proteins and the Arp2/3 complex, crucial for actin branching, may diminish the metastatic potential of cancer cells.

Besides this, the modulation of the immune response within the tumour microenvironment by WASP/WAVE

proteins is of significant importance. By targeting these proteins, it might be possible to enhance the effectiveness of immunotherapies, altering the behaviour of immune cells such as macrophages and potentially fostering an anti-tumorigenic milieu.

However, the journey to develop such treatments is laden with obstacles, especially regarding the specificity and safety of potential therapies. It is imperative to devise interventions that selectively target WASP/WAVE proteins in cancer cells while maintaining their essential roles in normal cells to avoid negative side effects. Additionally, the complex nature of the tumour microenvironment, heavily influenced by WASP/WAVE proteins, calls for extensive research, it can help in devising therapeutic strategies that can effectively interrupt these dynamics without unintentionally aiding tumour advancement.

Conflicts of Interest

The Authors declare no conflicts of interest. This research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Authors' Contributions

Wen G. Jiang and Tracey A. Martin conceptualized the research theme and were involved in all stages of the drafting. Rhiannon Yannan Yu performed the literature search, laid out the structure of the review, and wrote the initial draft of the manuscript. Subsequent revisions and the final preparation of the manuscript for submission were also carried out by Rhiannon Yannan Yu. Tracey A. Martin proofread and revised the manuscript. Wen G. Jiang read and approved the final manuscript.

Acknowledgements

I would like to express deep gratitude to my supervisor, Dr. Tracey A. Martin, whose insights and guidance were crucial at every stage of this research. Her unwavering support and encouragement were invaluable throughout the writing process. Prof. Wen G. Jiang provided not only expert advice but also substantial encouragement and support throughout this study. His extensive knowledge and insightful comments significantly contributed to the refinement of this review.

References

1 Siegel RL, Giaquinto AN, Jemal A: Cancer statistics, 2024. CA Cancer J Clin 74(1): 12-49, 2024. DOI: 10.3322/caac.21820

2 Chhikara BS, Parang K: Global Cancer Statistics 2022: the trends projection analysis. Chem Biol Lett 10(1): 451, 2023.

3 Steeg PS: Tumor metastasis: mechanistic insights and clinical challenges. Nat Med 12(8): 895-904, 2006. DOI: 10.1038/nm1469

4 Ridley AJ, Schwartz MA, Burridge K, Firtel RA, Ginsberg MH, Borisy G, Parsons JT, Horwitz AR: Cell migration: integrating signals from front to back. Science 302(5651): 1704-1709, 2003. DOI: 10.1126/science.1092053

5 Ridley AJ: Life at the leading edge. Cell 145(7): 1012-1022, 2011. DOI: 10.1016/j.cell.2011.06.010

6 Insall RH, Machesky LM: Actin dynamics at the leading edge: from simple machinery to complex networks. Dev Cell 17(3): 310-322, 2009. DOI: 10.1016/j.devcel.2009.08.012

7 Raftopoulou M, Hall A: Cell migration: Rho GTPases lead the way. Dev Biol 265(1): 23-32, 2004. DOI: 10.1016/j.ydbio.2003.06.003

8 Takenawa T, Suetsugu S: The WASP–WAVE protein network: connecting the membrane to the cytoskeleton. Nat Rev Mol Cell Biol 8(1): 37-48, 2007. DOI: 10.1038/nrm2069

9 Iwaya K, Norio K, Mukai K: Coexpression of Arp2 and WAVE2 predicts poor outcome in invasive breast carcinoma. Mod Pathol 20(3): 339-343, 2007. DOI: 10.1038/modpathol.3800741

10 Frugtniet B, Jiang WG, Martin TA: Role of the WASP and WAVE family proteins in breast cancer invasion and metastasis. Breast Cancer (Dove Med Press) 7: 99-109, 2015. DOI: 10.2147/BCTT.S59006

11 Yamaguchi H, Condeelis J: Regulation of the actin cytoskeleton in cancer cell migration and invasion. Biochim Biophys Acta 1773(5): 642-652, 2007. DOI: 10.1016/j.bbamcr.2006.07.001

12 Derry JM, Ochs HD, Francke U: Isolation of a novel gene mutated in Wiskott-Aldrich syndrome. Cell 78(4): 635-644, 1994. DOI: 10.1016/0092-8674(94)90528-2

13 Stewart DM, Treiber-Held S, Kurman CC, Facchetti F, Notarangelo LD, Nelson DL: Studies of the expression of the Wiskott-Aldrich syndrome protein. J Clin Invest 97(11): 2627-2634, 1996. DOI: 10.1172/JCI118712

14 Miki H, Miura K, Takenawa T: N-WASP, a novel actin-depolymerizing protein, regulates the cortical cytoskeletal rearrangement in a PIP2-dependent manner downstream of tyrosine kinases. EMBO J 15(19): 5326-5335, 1996. DOI: 10.1002/j.1460-2075.1996.tb00917.x

15 Miki H, Suetsugu S, Takenawa T: WAVE, a novel WASP-family protein involved in actin reorganization induced by Rac. EMBO J 17(23): 6932-6941, 1998. DOI: 10.1093/emboj/17.23.6932

16 Oda A, Eto K: WASPs and WAVEs: from molecular function to physiology in hematopoietic cells. Semin Cell Dev Biol 24(4): 308-313, 2013. DOI: 10.1016/j.semcdb.2013.03.002

17 Pollitt AY, Insall RH: WASP and SCAR/WAVE proteins: the drivers of actin assembly. J Cell Sci 122(Pt 15): 2575-2578, 2009. DOI: 10.1242/jcs.023879

18 Zhang J, Dong B, Siminovitch KA: Contributions of Wiskott–Aldrich syndrome family cytoskeletal regulatory adapters to immune regulation. Immunol Rev 232(1): 175-194, 2009. DOI: 10.1111/j.1600-065x.2009.00846.x

19 Miki H, Takenawa T: Regulation of actin dynamics by WASP family proteins. J Biochem 134(3): 309-313, 2003. DOI: 10.1093/jb/mvg146

20 Kurisu S, Takenawa T: The WASP and WAVE family proteins. Genome Biol 10(6): 226, 2009. DOI: 10.1186/gb-2009-10-6-226

21 Ho HY, Rohatgi R, Lebensohn AM, Le Ma, Li J, Gygi SP, Kirschner MW: Toca-1 mediates Cdc42-dependent actin nucleation by activating the N-WASP-WIP complex. Cell 118(2): 203-216, 2004. DOI: 10.1016/j.cell.2004.06.027

22 Rohatgi R, Nollau P, Ho HH, Kirschner MW, Mayer BJ: Nck and phosphatidylinositol 4,5-bisphosphate synergistically activate actin polymerization through the N-WASP-Arp2/3 pathway. J Biol Chem 276(28): 26448-26452, 2001. DOI: 10.1074/jbc.m103856200

23 Panchal SC, Kaiser DA, Torres E, Pollard TD, Rosen MK: A conserved amphipathic helix in WASP/Scar proteins is essential for activation of Arp2/3 complex. Nat Struct Biol 10(8): 591-598, 2003. DOI: 10.1038/nsb952

24 Yamaguchi H, Miki H, Suetsugu S, Ma L, Kirschner MW, Takenawa T: Two tandem verprolin homology domains are necessary for a strong activation of Arp2/3 complex-induced actin polymerization and induction of microspike formation by N-WASP. Proc Natl Acad Sci USA 97(23): 12631-12636, 2000. DOI: 10.1073/pnas.190351397

25 Takenawa T: From N-WASP to WAVE: Key molecules for regulation of cortical actin organization. Methods Enzymol 269: 3-15, 2005. DOI: 10.1002/047001766x.ch2

26 Vartiainen MK, Machesky LM: The WASP–Arp2/3 pathway: genetic insights. Curr Opin Cell Biol 16(2): 174-181, 2004. DOI: 10.1016/j.ceb.2004.02.004

27 Padrick SB, Rosen MK: Physical mechanisms of signal integration by WASP family proteins. Annu Rev Biochem 79: 707-735, 2010. DOI: 10.1146/annurev.biochem.77.06 0407.135452

28 Stradal TE, Rottner K, Disanza A, Confalonieri S, Innocenti M, Scita G: Regulation of actin dynamics by WASP and WAVE family proteins. Trends Cell Biol 14(6): 303-311, 2004. DOI: 10.1016/j.tcb.2004.04.007

29 Moreau V, Frischknecht F, Reckmann I, Vincentelli R, Rabut G, Stewart D, Way M: A complex of N-WASP and WIP integrates signalling cascades that lead to actin polymerization. Nat Cell Biol 2(7): 441-448, 2000. DOI: 10.1038/35017080

30 Rottner K, Stradal TE: Actin dynamics and turnover in cell motility. Curr Opin Cell Biol 23(5): 569-578, 2011. DOI: 10.1016/j.ceb.2011.07.003

31 Chen B, Chou HT, Brautigam CA, Xing W, Yang S, Henry L, Doolittle LK, Walz T, Rosen MK: Rac1 GTPase activates the WAVE regulatory complex through two distinct binding sites. Elife 6: e29795, 2017. DOI: 10.7554/eLife.29795

32 Ding B, Yang S, Schaks M, Liu Y, Brown AJ, Rottner K, Chowdhury S, Chen B: Structures reveal a key mechanism of WAVE regulatory complex activation by Rac1 GTPase. Nat Commun 13(1): 5444, 2022. DOI: 10.1038/s41467-022-33174-3

33 Lane J, Martin T, Weeks HP, Jiang WG: Structure and role of WASP and WAVE in Rho GTPase signalling in cancer. Cancer Genomics Proteomics 11(3): 155-165, 2014.

34 Derivery E, Gautreau A: Generation of branched actin networks: assembly and regulation of the N-WASP and WAVE molecular machines. Bioessays 32(2): 119-131, 2010. DOI: 10.1002/bies.200900123

35 Eden S, Rohatgi R, Podtelejnikov AV, Mann M, Kirschner MW: Mechanism of regulation of WAVE1-induced actin nucleation by Rac1 and Nck. Nature 418(6899): 790-793, 2002. DOI: 10.1038/nature00859

36 Winder SJ, Ayscough KR: Actin-binding proteins. J Cell Sci 118(4): 651-654, 2005. DOI: 10.1242/jcs.01670

37 Pollard TD, Cooper JA: Actin, a central player in cell shape and movement. Science 326(5957): 1208-1212, 2009. DOI: 10.1126/science.1175862

38 Davidson AJ, Wood W: Unravelling the actin cytoskeleton: a new competitive edge? Trends Cell Biol 26(8): 569-576, 2016. DOI: 10.1016/j.tcb.2016.04.001

39 Weeks HP, Sanders AJ, Kynaston HG, Jiang WG: The association between WAVE1 and -3 and the ARP2/3 complex in PC3 cells. Anticancer Res 36(3): 1135-1142, 2016.

40 Parker KK, Brock AL, Brangwynne C, Mannix RJ, Wang N, Ostuni E, Geisse NA, Adams JC, Whitesides GM, Ingber DE: Directional control of lamellipodia extension by constraining cell shape and orienting cell tractional forces. FASEB J 16(10): 1195-1204, 2002. DOI: 10.1096/fj.02-0038com

41 Le Clainche C, Carlier MF: Regulation of actin assembly associated with protrusion and adhesion in cell migration. Physiol Rev 88(2): 489-513, 2008. DOI: 10.1152/physrev.00021.2007

42 Krause M, Gautreau A: Steering cell migration: lamellipodium dynamics and the regulation of directional persistence. Nat Rev Mol Cell Biol 15(9): 577-590, 2014. DOI: 10.1038/nrm3861

43 Mattila PK, Lappalainen P: Filopodia: molecular architecture and cellular functions. Nat Rev Mol Cell Biol 9(6): 446-454, 2008. DOI: 10.1038/nrm2406

44 Pollard TD, Borisy GG: Cellular motility driven by assembly and disassembly of actin filaments. Cell 112(4): 453-465, 2003. DOI: 10.1016/s0092-8674(03)00120-x

45 Small JV, Stradal T, Vignal E, Rottner K: The lamellipodium: where motility begins. Trends Cell Biol 12(3): 112-120, 2002. DOI: 10.1016/s0962-8924(01)02237-1

46 Vignjevic DM, Montagnac G: Reorganisation of the dendritic actin network during cancer cell migration and invasion. Semin Cancer Biol 18(1): 12-22, 2008. DOI: 10.1016/j.semcancer.2007.08.001

47 Flanagan LA, Chou J, Falet H, Neujahr R, Hartwig JH, Stossel TP: Filamin A, the Arp2/3 complex, and the morphology and function of cortical actin filaments in human melanoma cells. J Cell Biol 155(4): 511-517, 2001. DOI: 10.1083/jcb.200105148

48 Rouiller I, Xu XP, Amann KJ, Egile C, Nickell S, Nicastro D, Li R, Pollard TD, Volkmann N, Hanein D: The structural basis of actin filament branching by the Arp2/3 complex. J Cell Biol 180(5): 887-895, 2008. DOI: 10.1083/jcb.200709092

49 Faix J, Rottner K: The making of filopodia. Curr Opin Cell Biol 18(1): 18-25, 2006. DOI: 10.1016/j.ceb.2005.11.002

50 Hahne P, Sechi AS, Benesch S, Small JV: Scar/WAVE is localised at the tips of protruding lamellipodia in living cells. FEBS Lett 492(3): 215-220, 2001. DOI: 10.1016/s0014-5793(01)02239-6

51 Kurisu S, Takenawa T: WASP and WAVE family proteins: friends or foes in cancer invasion? Cancer Sci 101(10): 2093-2104, 2010. DOI: 10.1111/j.1349-7006.2010.01654.x

52 Rotty JD, Wu CH, Bear JE: New insights into the regulation and cellular functions of the ARP2/3 complex. Nat Rev Mol Cell Biol 14(1): 7-12, 2013. DOI: 10.1038/nrm3492

53 Yamazaki D, Kurisu S, Takenawa T: Regulation of cancer cell motility through actin reorganization. Cancer Sci 96(7): 379-386, 2005. DOI: 10.1111/j.1349-7006.2005.00062.x

54 Fernando H, Kynaston H, Jiang WG: WASP and WAVE proteins: Vital intrinsic regulators of cell motility and their role in cancer (Review). Int J Mol Med 23(2): 141-148, 1998. DOI: 10.3892/ijmm_00000111

55 Friedl P, Wolf K: Tumour-cell invasion and migration: diversity and escape mechanisms. Nat Rev Cancer 3(5): 362-374, 2003. DOI: 10.1038/nrc1075

56 Tang H, Li A, Bi J, Veltman DM, Zech T, Spence HJ, Yu X, Timpson P, Insall RH, Frame MC, Machesky LM: Loss of Scar/WAVE complex promotes N-WASP- and FAK-dependent invasion. Curr Biol 23(2): 107-117, 2013. DOI: 10.1016/j.cub.2012.11.059

57 Yamaguchi H, Oikawa T: Membrane lipids in invadopodia and podosomes: key structures for cancer invasion and metastasis. Oncotarget 1(5): 320-328, 2010. DOI: 10.18632/oncotarget.164

58 McNiven MA, Baldassarre M, Buccione R: The role of dynamin in the assembly and function of podosomes and invadopodia. Front Biosci 9(1-3): 1944, 2004. DOI: 10.2741/1348

59 Artym VV, Zhang Y, Seillier-Moiseiwitsch F, Yamada KM, Mueller SC: Dynamic interactions of cortactin and membrane type 1 matrix metalloproteinase at invadopodia: Defining the stages of invadopodia formation and function. Cancer Res 66(6): 3034-3043, 2006. DOI: 10.1158/0008-5472.can-05-2177

60 Meirson T, Gil-Henn H: Targeting invadopodia for blocking breast cancer metastasis. Drug Resist Updat 39: 1-17, 2018. DOI: 10.1016/j.drup.2018.05.002

61 Artym VV, Matsumoto K, Mueller SC, Yamada KM: Dynamic membrane remodeling at invadopodia differentiates invadopodia from podosomes. Eur J Cell Biol 90(2-3): 172-180, 2011. DOI: 10.1016/j.ejcb.2010.06.006

62 Eddy RJ, Weidmann MD, Sharma VP, Condeelis JS: Tumor Cell Invadopodia: Invasive Protrusions that Orchestrate Metastasis. Trends Cell Biol 27(8): 595-607, 2017. DOI: 10.1016/j.tcb.2017.03.003

63 Stylli SS, Kaye AH, Lock P: Invadopodia: At the cutting edge of tumour invasion. J Clin Neurosci 15(7): 725-737, 2008. DOI: 10.1016/j.jocn.2008.03.003

64 Helgeson LA, Prendergast JG, Wagner AR, Rodnick-Smith M, Nolen BJ: Interactions with actin monomers, actin filaments, and Arp2/3 complex define the roles of WASP family proteins and cortactin in coordinately regulating branched actin networks. J Biol Chem 289(42): 28856-28869, 2014. DOI: 10.1074/jbc.M114.587527

65 Yamaguchi H, Lorenz M, Kempiak S, Sarmiento C, Coniglio S, Symons M, Segall J, Eddy R, Miki H, Takenawa T, Condeelis J: Molecular mechanisms of invadopodium formation: the role of the N-WASP-Arp2/3 complex pathway and cofilin. J Cell Biol 168(3): 441-452, 2005. DOI: 10.1083/jcb.200407076

66 Takenawa T, Miki H: WASP and WAVE family proteins: key molecules for rapid rearrangement of cortical actin filaments and cell mov. J Cell Sci 114(10): 1801-1809, 2001. DOI: 10.1242/jcs.114.10.1801

67 Suetsugu S, Yamazaki D, Kurisu S, Takenawa T: Differential roles of WAVE1 and WAVE2 in dorsal and peripheral ruffle formation for fibroblast cell migration. Dev Cell 5(4): 595-609, 2003. DOI: 10.1016/s1534-5807(03)00297-1

68 Pichot CS, Arvanitis C, Hartig SM, Jensen SA, Bechill J, Marzouk S, Yu J, Frost JA, Corey SJ: Cdc42-interacting protein 4 promotes breast cancer cell invasion and formation of invadopodia through activation of N-WASp. Cancer Res 70(21): 8347-8356, 2010. DOI: 10.1158/0008-5472.CAN-09-4149

69 Oser M, Yamaguchi H, Mader CC, Bravo-Cordero JJ, Arias M, Chen X, Desmarais V, van Rheenen J, Koleske AJ, Condeelis J: Cortactin regulates cofilin and N-WASp activities to control the stages of invadopodium assembly and maturation. J Cell Biol 186(4): 571-587, 2009. DOI: 10.1083/jcb.200812176
70 Lomakina ME, Lallemand F, Vacher S, Molinie N, Dang I, Cacheux W, Chipysheva TA, Ermilova VD, de Koning L, Dubois T, Bièche I, Alexandrova AY, Gautreau A: Arpin downregulation in breast cancer is associated with poor prognosis. Br J Cancer 114(5): 545-553, 2016. DOI: 10.1038/bjc.2016.18
71 Kazazian K, Go C, Wu H, Brashavitskaya O, Xu R, Dennis JW, Gingras AC, Swallow CJ: Plk4 promotes cancer invasion and metastasis through Arp2/3 complex regulation of the actin cytoskeleton. Cancer Res 77(2): 434-447, 2017. DOI: 10.1158/0008-5472.can-16-2060
72 Laporte S, Magistrato A: Deciphering the molecular terms of Arp2/3 allosteric regulation from all-atom simulations and dynamical network theory. J Phys Chem Lett 12(22): 5384-5389, 2021. DOI: 10.1021/acs.jpclett.1c00940
73 Yamaguchi H: Pathological roles of invadopodia in cancer invasion and metastasis. Eur J Cell Biol 91(11-12): 902-907, 2012. DOI: 10.1016/j.ejcb.2012.04.005
74 Zicha D, Allen WE, Brickell PM, Kinnon C, Dunn GA, Jones GE, Thrasher AJ: Chemotaxis of macrophages is abolished in the Wiskott-Aldrich syndrome. Br J Haematol 101(4): 659-665, 1998. DOI: 10.1046/j.1365-2141.1998.00767.x
75 Ishihara D, Dovas A, Hernandez L, Pozzuto M, Wyckoff J, Segall JE, Condeelis JS, Bresnick AR, Cox D: Wiskott-Aldrich syndrome protein regulates leukocyte-dependent breast cancer metastasis. Cell Rep 4(3): 429-436, 2013. DOI: 10.1016/j.celrep.2013.07.007
76 Martin TA, Pereira G, Watkins G, Mansel RE, Jiang WG: N-WASP is a putative tumour suppressor in breast cancer cells, *in vitro* and *in vivo*, and is associated with clinical outcome in patients with breast cancer. Clin Exp Metastasis 25(2): 97-108, 2008. DOI: 10.1007/s10585-007-9120-8
77 Escudero-Esparza A, Jiang WG, Martin TA: Claudin-5 is involved in breast cancer cell motility through the N-WASP and ROCK signalling pathways. J Exp Clin Cancer Res 31(1): 43, 2012. DOI: 10.1186/1756-9966-31-43
78 Meirovitz A, Nisman B, Allweis TM, Carmon E, Kadouri L, Maly B, Maimon O, Peretz T: Thyroid hormones and morphological features of primary breast cancer. Anticancer Res 42(1): 253-261, 2022. DOI: 10.21873/anticanres.15480
79 Gligorijevic B, Wyckoff J, Yamaguchi H, Wang Y, Roussos ET, Condeelis J: N-WASP-mediated invadopodium formation is involved in intravasation and lung metastasis of mammary tumors. J Cell Sci 125(Pt 3): 724-734, 2012. DOI: 10.1242/jcs.092726
80 Hebbrecht T, Van Audenhove I, Zwaenepoel O, Verhelle A, Gettemans J: VCA nanobodies target N-WASp to reduce invadopodium formation and functioning. PLoS One 12(9): e0185076, 2017. DOI: 10.1371/journal.pone.0185076
81 Chung YJ, Salvi A, Kalailingam P, Alnawaz M, Tan SH, Pan JY, Tan NS, Thanabalu T: N-WASP attenuates cell proliferation and migration through ERK2-dependent enhanced expression of TXNIP. Biology (Basel) 11(4): 582, 2022. DOI: 10.3390/biology11040582
82 Al Absi A, Wurzer H, Guerin C, Hoffmann C, Moreau F, Mao X, Brown-Clay J, Petrolli R, Casellas CP, Dieterle M, Thiery JP, Chouaib S, Berchem G, Janji B, Thomas C: Actin cytoskeleton remodeling drives breast cancer cell escape from natural killer-mediated cytotoxicity. Cancer Res 78(19): 5631-5643, 2018. DOI: 10.1158/0008-5472.can-18-0441
83 Mondaca JM, Uzair ID, Castro Guijarro AC, Flamini MI, Sanchez AM: Molecular basis of LH action on breast cancer cell migration and invasion *via* kinase and scaffold proteins. Front Cell Dev Biol 8: 630147, 2021. DOI: 10.3389/fcell.2020.630147
84 Shortrede JE, Uzair ID, Neira FJ, Flamini MI, Sanchez AM: Paxillin, a novel controller in the signaling of estrogen to FAK/N-WASP/Arp2/3 complex in breast cancer cells. Mol Cell Endocrinol 430: 56-67, 2016. DOI: 10.1016/j.mce.2016.04.007
85 Guerriero CJ, Weisz OA: N-WASP inhibitor wiskostatin nonselectively perturbs membrane transport by decreasing cellular ATP levels. Am J Physiol Cell Physiol 292(4): C1562-C1566, 2007. DOI: 10.1152/ajpcell.00426.2006
86 Fernando HS, Davies SR, Chhabra A, Watkins G, Douglas-Jones A, Kynaston H, Mansel RE, Jiang WG: Expression of the WASP Verprolin-Homologues (WAVE members) in human breast cancer. Int J Oncol 31(5-6): 376-383, 2007. DOI: 10.1159/000136157
87 Koike E, Iwaya K, Watanabe A, Miyake S, Sato E, Ishikawa T: Association between breast cancer recurrence and cellular dissociation assessed using fine-needle aspiration. Cytopathology 60(5): 413-420, 2016. DOI: 10.1159/000448508
88 Ko HS, Kim JS, Cho SM, Lee HJ, Ahn KS, Kim SH, Lee EO: Urokinase-type plasminogen activator expression and Rac1/WAVE-2/Arp2/3 pathway are blocked by pterostilbene to suppress cell migration and invasion in MDA-MB-231 cells. Bioorg Med Chem Lett 24(4): 1176-1179, 2014. DOI: 10.1016/j.bmcl.2013.12.115
89 Algayadh IG, Dronamraju V, Sylvester PW: Role of Rac1/WAVE2 signaling in mediating the inhibitory effects of γ-tocotrienol on mammary cancer cell migration and invasion. Biol Pharm Bull 39(12): 1974-1982, 2016. DOI: 10.1248/bpb.b16-00461
90 Kim JS, Kang CG, Kim SH, Lee E: Rhapontigenin suppresses cell migration and invasion by inhibiting the PI3K-dependent Rac1 signaling pathway in MDA-MB-231 human breast cancer cells. J Nat Prod 77(5): 1135-1139, 2014. DOI: 10.1021/np401078g

91 Rana PS, Wang W, Markovic V, Szpendyk J, Chan ER, Sossey-Alaoui K: The WAVE2/miR-29/Integrin-β1 oncogenic signaling axis promotes tumor growth and metastasis in triple-negative breast cancer. Cancer Res Commun 3(1): 160-174, 2023. DOI: 10.1158/2767-9764.CRC-22-0249
92 Yamashita H, Ueda K, Kioka N: WAVE2 forms a complex with PKA and is involved in PKA enhancement of membrane protrusions. J Biol Chem 286(5): 3907-3914, 2011. DOI: 10.1074/jbc.M110.145409
93 Rana PS, Alkrekshi A, Wang W, Markovic V, Sossey-Alaoui K: The role of WAVE2 signaling in cancer. Biomedicines 9(9): 1217, 2021. DOI: 10.3390/biomedicines9091217
94 Sossey-Alaoui K, Ranalli TA, Li X, Bakin AV, Cowell JK: WAVE3 promotes cell motility and invasion through the regulation of MMP-1, MMP-3, and MMP-9 expression. Exp Cell Res 308(1): 135-145, 2005. DOI: 10.1016/j.yexcr.2005.04.011
95 Sossey-Alaoui K, Safina A, Li X, Vaughan MM, Hicks DG, Bakin AV, Cowell JK: Down-regulation of WAVE3, a metastasis promoter gene, inhibits invasion and metastasis of breast cancer cells. Am J Pathol 170(6): 2112-2121, 2007. DOI: 10.2353/ajpath.2007.060975
96 Sossey-Alaoui K, Li X, Cowell JK: c-Abl-mediated phosphorylation of WAVE3 is required for lamellipodia formation and cell migration. J Biol Chem 282(36): 26257-26265, 2007. DOI: 10.1074/jbc.m701484200
97 Sossey-Alaoui K, Bialkowska K, Plow EF: The miR200 family of microRNAs regulates WAVE3-dependent cancer cell invasion. J Biol Chem 284(48): 33019-33029, 2009. DOI: 10.1074/jbc.M109.034553
98 Sossey-Alaoui K, Downs-Kelly E, Das M, Izem L, Tubbs R, Plow EF: WAVE3, an actin remodeling protein, is regulated by the metastasis suppressor microRNA, miR-31, during the invasion-metastasis cascade. Int J Cancer 129(6): 1331-1343, 2011. DOI: 10.1002/ijc.25793
99 Sossey-Alaoui K: Surfing the big WAVE: Insights into the role of WAVE3 as a driving force in cancer progression and metastasis. Semin Cell Dev Biol 24(4): 287-297, 2013. DOI: 10.1016/j.semcdb.2012.10.006
100 Kulkarni S, Augoff K, Rivera L, McCue B, Khoury T, Groman A, Zhang L, Tian L, Sossey-Alaoui K: Increased expression levels of WAVE3 are associated with the progression and metastasis of triple negative breast cancer. PLoS One 7(8): e42895, 2012. DOI: 10.1371/journal.pone.0042895
101 Taylor MA, Davuluri G, Parvani JG, Schiemann BJ, Wendt MK, Plow EF, Schiemann WP, Sossey-Alaoui K: Upregulated WAVE3 expression is essential for TGF-β-mediated EMT and metastasis of triple-negative breast cancer cells. Breast Cancer Res Treat 142(2): 341-353, 2013. DOI: 10.1007/s10549-013-2753-1
102 Spence HJ, Timpson P, Tang HR, Insall RH, Machesky LM: Scar/WAVE3 contributes to motility and plasticity of lamellipodial dynamics but not invasion in three dimensions. Biochem J 448(1): 35-42, 2012. DOI: 10.1042/BJ20112206
103 Wang W, Kansakar U, Markovic V, Wang B, Sossey-Alaoui K: WAVE3 phosphorylation regulates the interplay between PI3K, TGF-β, and EGF signaling pathways in breast cancer. Oncogenesis 9(10): 87, 2020. DOI: 10.1038/s41389-020-00272-0
104 Kansakar U, Wang W, Markovic V, Sossey-Alaoui K: Phosphorylation of the proline-rich domain of WAVE3 drives its oncogenic activity in breast cancer. Sci Rep 11(1): 3868, 2021. DOI: 10.1038/s41598-021-83479-4
105 Kansakar U, Wang W, Markovic V, Sossey-Alaoui K: Elucidating the molecular signaling pathways of WAVE3. Ann Transl Med 8(14): 900, 2020. DOI: 10.21037/atm.2020.02.16
106 Davuluri G, Schiemann WP, Plow EF, Sossey-Alaoui K: Loss of WAVE3 sensitizes triple-negative breast cancers to chemotherapeutics by inhibiting the STAT-HIF-1α-mediated angiogenesis. JAKSTAT 3(4): e1009276, 2015. DOI: 10.1080/21623996.2015.1009276
107 Wang W, Rana PS, Markovic V, Sossey-Alaoui K: The WAVE3/β-catenin oncogenic signaling regulates chemoresistance in triple negative breast cancer. Breast Cancer Res 25(1): 31, 2023. DOI: 10.1186/s13058-023-01634-3
108 Qasim M, Ricks-Santi LJ, Naab TJ, Rajack F, Beyene D, Abbas M, Kassim OO, Copeland RL, Kanaan Y: Inverse correlation of KISS1 and KISS1R expression in triple-negative breast carcinomas from African American women. Cancer Genomics Proteomics 19(6): 673-682, 2022. DOI: 10.21873/cgp.20350