A review on the chemistry and pharmacological properties of benzodiazepine motifs in drug design

Olayinka O. Tolu-Bolajia, Samuel O. Sojinu, Adebola P. Okederea and Olayinka O. Ajani

ABSTRACT

Benzodiazepines are an important class of heterocyclic compounds in organic chemistry. They are known for their diverse physicochemical and biological properties. Some benzodiazepine derivatives are well-known drugs with diverse and strong pharmacophoric moiety. An immense number of pharmacological research on benzodiazepine heterocycles and their derivatives have recently been conducted to explore its numerous pharmacological potentials as better therapeutic candidates for the treatment of various disorders, benzodiazepines, however, are one of the main sources of interest for many medicinal chemists.

1. Introduction

Heterocyclic compounds are essential group of medicinal compounds with more than half of the well-known organic medications contained this template (Ajani et al., 2019). Because of its significance in the study of pharmacological variety in medicinal chemistry and organic synthesis, it may be claimed that the world is currently in the phase of heterocyclic chemistry (Ajani et al., 2019). The foundation of planetary and human existence is heterocyclic chemistry which is a large subset of organic compounds with a diversity of biological functions (Balaban, Onciu, & Katritzky, 2004).

For the quantification and confirmation of biologically active heterocyclic based medicines, spectrophotometric analysis has been recognized as the essential toolbox (Azmi et al., 2013; Azmi, Al-Fazari, Al-Badael, & Al-Mahrazi, 2015; Azmi, Al-Hadhrami, Al-Marhoubi, Al-Sulaimi, & Al-Shamoosi, 2017). For instance, imipramine hydrochloride, a benzodiazepine biosostere, has just been validated in tablet as solid material by the use of spectrophotometric determination (Azmi et al., 2022). Additionally, the detection of doxepin hydrochloride in commercial dosage forms as well as the identification of piroxicam in commercial dosage forms have both been effectively accomplished using the spectrofluorimetric approach (Azmi, Iqbal, Jaboob, Al Shahari, & Rahman, 2009; Rahman, Siddiqui, & Azmi, 2009). In order to conclude all investigations and acquire a suitable specification to ensure the level of purity of drug substances and drug products, a proposed method of profiling drug impurity has been made (Rahman, Azmi, & Wu, 2006).

Benzodiazepine, a potent pharmacophore of crucial biodiversity for drug discovery, is the heterocyclic molecule of focus in this study. The psychoactive substance benzodiazepine, and its derivatives are bicyclic heterocyclic compounds having a benzene ring fused to a seven-membered ring. The diazepine ring has two nitrogen atoms at various places in the ring (Shorter, 2005). Regardless of where the nitrogen atoms are located, benzodiazepines are numbered from the position next to the carbocyclic ring. Hoffmann-La Roche discovered the first benzodiazepine in 1955, and Librium, its brand name, was introduced to consumers in 1960 (Wicy, 2013).

Benzodiazepine motifs are found in trace amount in Carob plants (Ceratonia siliqua) and are used as anxiolytics, hypnotics and as chemo-anticipatory agents (Avallone, Cosenza, Farina, Baraldi, & Baraldi, 2002).
They are well-known in treating insomnia, anxiety, agitation and depression as well as being anti-epilepsy (Mbomo, Omam, Kavaye, Kameni, & Bum, 2015). Benzodiazepines can be found in *Mimosa pudica* (touch me not leaves). In addition, they can also be extracted from biological fluid (Westland & Dorman, 2013) and *Erythrina velutina* Willd (Fabaceae plant) (Danta et al., 2004; Ribeiro, Onusic, Poltronieri, & Viana, 2006; Teixeira-Silva et al., 2008). However, benzodiazepine was first established in mammalian tissues in 1986 (Burman et al., 2019; Sand et al., 2000). The first chlordiazepoxide (benzodiazepine) was first revealed by Leo Sternbach and his team in 1957 while reviewing the action of quinazoline oxide (Wicy, 2013).

Furthermore, benzodiazepines are engaged as useful therapeutic agents (Dubovsky & Marshall, 2022). Some of the derivatives can serve as useful intermediates in multistep synthesis. Moreover, benzodiazepines can also be used as fine chemicals during drug design with diverse functionalities and better biological properties. The pharmacological activities exhibited by benzodiazepine are, but not limited to anti-depressant (Anderson, 2022; Ogawa et al., 2019; Sharma, Tilak, Thakur, Gangwar, & Sutar, 2017), anti-inflammatory (Ongone et al., 2019), antihypertensive (Wright et al., 2021), antimalarial (Dzierszinski et al., 2002; Insuasty et al., 2017), antitumor (Kim et al., 2021), anticancer (Chen et al., 2014; Misra et al., 2020), anticonvulsant (Gaponov et al., 2016; Shao, Han, Wu, & Piao, 2018), antibacterial (Verma et al., 2019), antitubercular (Anil et al., 2019; Kumar, Mohan, Mai, Sangeeta, & Nagasree, 2018), anti-proliferative (Sharp et al., 2017) and antifungal (Caiana, de Veras, de Souza, & Queiroz, 2021; Paulussen et al., 2014) activities.

However, due to diverse medicinal potential, drug ability and other applications, numerous derivatives of benzodiazepine have been synthesized in the laboratory. In most cases o-phenylenediamine was used as the precursor and it was reacted with aliphatic ketone indifferent catalysts such as H-MCM-22 (Majid, Khanday, & Tomar, 2012); p-toluenesulfonic acid (Pasha & Jayashankara, 2006); copper nano particle (Shaikh et al., 2020); palladium catalyst (Christodoulou, Beccalli, & Giofrè, 2020); iridium-catalyzed in bisphosphine support (Verzijl, Hassfeld, Hassfeld, de Vries, & Lefort, 2020); natural/synthetic zeolites (Perez-Mayoral & Lopez-Peinado, 2021); sulfated polyborate (Indalkar, Patil, & Chaturbhuj, 2017); glycerol (Radatz et al., 2011); while other involved reactions of the same precursor with alkyne (Qian, Liu, Cui, & Xu, 2012); isocyanomethyl benzene (Chen et al., 2014) and propargylic alcohol (Cacchi, Fabrizi, Goggiamani, & Iazzetti, 2016).

Based on the location of the nitrogen atom in the seven-membered rings, there are six significant benzodiazepine ring structures: 5H-1,2-benzodiazepine 1, 1H-1,3-benzodiazepine 2, 3H-1,4-benzodiazepine 3, 3H-1,5-benzodiazepine 4, 5H-2,3-benzodiazepine 5 and 1H-2,4-benzodiazepine 6 (Figure 1). Over 20 of the nearly 40 benzodiazepine drugs on the market have received global pharmacological approval (Soyka, 2017). A recent evaluation of various benzodiazepine-based chemical synthesis techniques placed particular emphasis on the multicomponent reaction as a useful toolbox (Farhid, Khodkari, Nazeri, Javanbakht, & Shaabani, 2021). The following commercially available benzodiazepine derivatives are beneficial drug candidates: Chlordiazepoxide 7 and Alprazolam 8 are prescription drugs for panic disorder. Clonazepam (9, used to control epilepsy), diazepam (10, for nerve and brain tranquility), lorazepam (11, for sleep disorders), temazepam (12), and quazepam 13 (Figure 2).

**Figure 1.** Six important ring structures of benzodiazepines in view of their nitrogen positions.
1.1. Justification for the review

There have been some recent reviews on benzodiazepine, for instance, da Silva et al. (2021) focussed their review on formation of organometallic complexes, Arora, Dhiman, Kumar, Singh, and Monga (2020) put emphases on recent advances in synthesis and medicinal chemistry, Christodoulou et al. (2020) centered their review on the synthesis of benzodiazepine via palladium-catalyst. Also, Farhid et al. (2021), focused their review on the multicomponent reaction as a vital instrument for the formation of benzodiazepines. Velasco-Rubio, Varela, and Saa (2020) reviewed mainly on the transition metals as catalysts in the production of benzazepines and benzodiazepines. Varvounis (2016) has his review mainly from 2010 to 2015. In addition, Rashid et al. (2019) centered their review on 1,4-diazepines formatiom, reaction and biological usefulness.

Meanwhile, the introduction of long-lasting synthetic techniques that isolate specified compounds in an efficient and environmentally responsible manner is the primary constraint facing the pharmacological business in synthetic organic chemistry. Due to benzodiazepines remarkable biological characteristics and success, the study of this system provides a very fascinating field for exploration in drug research. Therefore, this review includes a variety of plant extracts, a recent synthetic approach, and pharmacological properties to address the ongoing need to find improved medications that will deliver the highest level of clinical benefits. This present review also provides avenue to harness most recent updates after the publications date of those previous reviews so as to bridge necessary gaps.

2. Chemistry

Benzodiazepine and other heterocyclic-based templates are core components of drug design. They have been extracted in various form using diverse methods. The extraction method and the applications of some of these selected biomolecular entities is as shown in Table 1. For instance, Doxepin hydrochloride which is a tricyclic heterocyclic compound was extracted through fluorescent ion pair complex formation with eosin Y. It was effectively extracted with dichloromethane using buffer solution of pH 4.52 (Rahman et al., 2009). Piroxicam as a non-steroidal anti-inflammatory drug (NSAID) was spectrophotometrically determined after effective extraction in ethanol-water medium (Table 1). Method was based on the chelation of the drug with Fe(III) in ethanol-water medium to form a pink colored complex which absorbs maximally at 504 nm (Azmi et al., 2009). The liquid-liquid extraction approach was also used for detection of piroxicam in human plasma and subsequently analyzed with HPLC (Calvo et al., 2016).

Benzodiazepine based drugs have been extracted from diverse medicinal plants and their biomass feed stocks using various methods. However, benzodiazepine derivatives are found in trace amount in Carod plants (Ceatonia siliqua) and are used as
2.1. Synthesis

2.1.1. Synthesis via H-MCM-22 catalyzed approach

According to Majid et al. (2012), the reaction of a calculated amount of o-phenylenediamine, the matching ketone, and H-MCM-22 catalyst produced eight examples of 1,5-benzodiazepines derivatives (Scheme 1). Compound 14a-h was produced from acetone and used to demonstrate the catalyst’s recyclable nature. Its initial yield was 87%, while the yields obtained from its synthesis using H-three MCM-22’s time’s recyclable effort were 85%, 81%, and 75%, respectively. This demonstrated the great efficiency of recovery and reuse for this catalyst. In order to obtain 1,5-benzodiazepines with a very good yield, the combination was employed and reacted in the CH3CN at normal temperature and pressure. It was reported that H-MCM-22 catalyst was highly expeditious and efficient for both cyclic and acyclic ketones. Some shortcoming like corrosiveness, toxic nature, waste products generation, and expensiveness evident in homogenous catalysts, were cheaply overcome by the use of H-MCM-22 catalyst.

2.1.2. Synthesis via glycerol recyclable solvent approach

Radatz et al. (2011) synthesized eight trisubstituted benzodiazepines 15a-h by reacting o-phenylenediamine with excess of acetophenone and other aromatic and aliphatic ketones using glycerol as solvent. At 25 °C, the yield of 15a was very low but when the temperature was elevated to 90 °C for 4 hours, it gave the product in an excellent yield of 96% (Scheme 2). It was validated that the glycerol could be reused for about four times with no loss of potency. Although, seven of these products were obtained in excellent yield (70-96%); however, this solvent-dependent catalyst-free reaction was not well favoured when simplest symmetrical ketone (acetone) was use as it produced the corresponding benzodiazepine, 15b in low yield (45%) despite the efficacy of glycerol in the reaction optimization study.

2.1.3. Synthesis via CdCl2 mediated approach

The synthesis of a series of ten imino-based benzodiazepines, 16a-j was carried out via condensative reaction of 1,2-phenylenediamine with acetooacetic anhydride using CdCl2 as catalyst under heat and microwave irradiation techniques (Scheme 3). The products were obtained in good yields (52-68%) within 2 mins under microwave irradiation. Although, none of them was obtained in excellent yield but the presence of catalyst improved the yields when compared with reaction in the absence of CdCl2 as catalyst. Effort was made to compare this with reaction under convention heating method under reflux and it was noted that all reaction were...
also completed at < 1 h under reflux heating. It occurred using the blending of tetrahydrofuran and acetic acid combined solvent system (Scheme 3). The microwave reaction occurred in short reaction times (Ilango, Remya, & Ponnuswamy, 2013).

2.1.4. Synthesis via one-pot three-component reaction approach
An array of twenty new benzodiazepine derivatives, 17a-t were synthesized via a special multi-component reaction involving one-pot three-component reaction with the three components being diamino-benzene-3-carboxamide, meldrum's acid and isocyanomethylbenzene (Scheme 4). This multicomponent reaction safes time, but its limitation in this present design was that the 17a-t products were obtained in low yields (25-45%). This compound 17a having R = isopropyl, was obtained in 45% which was the highest yield among the series. Hence, there may be need to improve on the solvent through the use of deep eutectic solvent in order to enhance the product yields. These compounds were screening for anticancer potential and they displayed good antitumor activities against human notable carcinoma (Chen et al., 2014).

2.1.5. Synthesis via p-toluenesulfonic acid catalyzed approach
The synthesis of 2,3-dihydro-1H-1,5-benzodiazepine 18 was carried out via the condensation of 1,2-phenylenediamine with 2-pentanone using catalytic amount of p-TsOH at 80°C–85°C for about 20 min. (Scheme 5). Benzodiazepine 18 was accessed in 92% yield. Other aliphatic and cyclic ketones were also utilized to obtain other seven benzodiazepine derivatives in varying yields (70-92%) and reaction time of not more than 20 min (Pasha & Jayashankara, 2006). The research effort here which used p-toluenesulfonic acid catalyzed condition to produce the benzodiazepine, occurred within 20 min while a similar reaction in glycerol by Radatz et al. (2011) furnished the benzodiazepine product after 4 hours. Hence, role of p-toluenesulfonic acid in lowering the activation energy for a better kinetic is well commendable.

2.1.6. Synthesis via gold(I)-catalyzed approach
The formation of an array of twenty-four derivatives of 1,5-benzodiazepine 19 was successfully reported from the reaction of 1,2-phenylenediamine with alkynes at 60°C in chloroform for 6 hours under the influence of a catalytic amount of Gold (I) (Scheme 6). Efficacy of diverse gold-based catalysts were investigated. Also, 

![Scheme 1. Synthesis via catalytic performance of H-MCM-22](image1)

![Scheme 2. Synthesis of benzodiazepines using glycerol as solvent.](image2)
biphenyl)Cy2PAuNTf2 featured as the most efficient catalytic substrate that resulted in highest yield within 6 h. Interestingly, 5 mol% catalytic amount was used for the product to be accessed in good yield (Qian et al., 2012). Out of these twenty-four benzodiazepines, the first fourteen \(19a-n\) were obtained in 46-97% yields by starting from un-substituted o-phenylenediamine precursor while the remaining ten \(19o-x\) were accessed in 62-99% yields using substituted o-phenylenediamine. Although, this reaction was high atom economical in accessing the 24 derivatives, but when prop-1-yn-1-ylbenzene was used as the alkyne synthon the reaction failed woefully.

2.1.7. Synthesis via photo-redox catalyzed radical cascade approach

The formation of fluorinated pyrrolo[1,2-d]benzodiazepine derivatives by means of photo-redox catalyzed radical cascade reaction was made possible in inert condition created by the presence of argon and reaction time of 24 h (Scheme 7). Specifically, when \(R_1 = R_2 = H; X = C\) this led to the formation of five tetracyclic benzodiazepines \(20a-e\). Single electron transfer (SET) played a crucial role in the initiation of the photochemical reaction reported therein. The reaction environment was basified with triethylamine with dichloromethane being the solvent (Lian et al., 2019). Cyclization was successful, but the yields were low (below 50%), except for \(20e\) which was obtained in 62% yield, which was also the only derivative where substitution took place on the indole ring. Thus, there may be need for further search on yield improvement strategy. The sixth attempt as \(20f\) failed woefully as the cyclization to benzodiazepine was abortive when benzimidazole was used instead of indole which showed that...
second nitrogen heteroatom of benzimidazole might have played a crucial in this failure.

2.1.8. **Synthesis using gold-catalyzed domino reaction**

The synthesis of eighteen benzodiazepines 21a-r was harnessed from catalytic performance of gold catalyzed hydroamination of 1,2-phenylenediamine and propargylic alcohols having substituents at positions 2 and 4 respectively (Scheme 8). Each of the products formed centered on the activity of R1 and R2. The last compound of this series was a yellow compound 21r (R1 = Ph, R2 = H) which was obtained in 77% yield (Cacchi et al., 2016) while other was obtained in 32-72%. This reaction was reported to proceed well at 60°C and was maintained as the refluxing temperature (Cacchi et al., 2016). It was observed that the presence of strong electron donor at the terminal portion of acetylenic bond resulted in benzodiazepine yield enhancement.

2.1.9. **Synthesis via Brønsted catalyzed approach**

The effective performance of Brønsted catalysts (MIL/K-SO3H and MIL/Ks-CN) were carried out through condensation of 1,2-phenylenediamine and ketones at 50°C in lieu of the formation of substituted 1,4-benzodiazepines (Scheme 9). When nona-5-one is used, the product 22 was obtained in 83%. MIL/Ks-CN indicated better catalytic efficiency and activities when compared with MIL/K-SO3H (Isaeva et al., 2019).

2.1.10. **Synthesis via environmental-friendly approach**

Indalkar et al. (2017) synthesized 4-substituted 1,4-benzodiazepine in an environment friendly one pot system through 3 components reaction which was effectively catalyzed by sulfated polyborate (Scheme 10). Optimization study for 23 productions was investigated using the reaction of o-phenylenediamine, cyclohexa-1,3-dione and un-substituted benzaldehyde. Effect of catalyst loading and temperature showed that the highest yield of 23 (95%) was obtained when 15 mol% of the catalyst was used at 100°C reaction temperature. The reaction time was short with excellent yield when the refluxing temperature was elevated to 100°C (Indalkar et al., 2017).

2.1.11. **Synthesis via copper catalyzed thermal cyclization**

Chen et al. (2020) produced 1,4-benzodiazepines-5-one, 24 (when R1 = R2 = R3 = R4 = R5 = H) by
copper catalyzed thermal cyclization of alkenylated anthranilamide derivatives (Scheme 11). This was mechanistically described to have occurred through copper-catalyzed rearrangement cascade allyl-amination and C=C rearrangement with good yield of not less than 90%. The reaction was completed in 12 h when the heating was done at 110 °C using toluene as solvent.

2.1.12. Synthesis via clay backing supported microwave approach
Shaikh et al. (2020) stated that microwave assisted preparation of thiophenyl-1,5-benzo diazepines were made possible by utilizing various clay backing catalyst and clay backing transition metals support (Scheme 12). Microwave assisted reaction of o-PDA with 3-acetyl thiophene using Cu(II)-clay nano catalyst afforded benzodiazepine 25 in 98% yield. Among all the metal clay catalysts, copper (II) on clay nano catalyst adsorbent was the most viable and exhibited excellent action with high yield based on the reported optimization study (Shaikh et al., 2020).

2.1.13. Synthesis from chalcones and 1,2-phenylenediamine
Taha and Rasheed (2022) reported the microwave assisted preparation of 1,5-benzodiazepine 26 via the condensation of 1,2-phenylenediamine on chalcone in absolute ethanol and NaOH (10% w/v). The chalcone utilized was prepared from the condensation reaction of acetylacetone with cinnamaldehyde in a basified environment (Scheme 13). This reaction proceeded smoothly in the microwave at 400-Watt power modulation (Taha & Rasheed, 2022).

2.1.14. Synthesis from AlKIT-5 catalysts at room
Shobha et al. (2010) stated that ambient temperature synthesis of 1,5-benzodiazepines was achieved by the use of various clay backing catalyst and cage type mesoporous aluminosilicate catalysts. When cyclopentanone was treated with o-phenylenediamine, 27 were obtained in 92% within 1 h reaction time (Scheme 14). It was reported that the high catalytic activity was due its high acidity and excellent textual parameters (Shobha et al., 2010).
3. Pharmacological properties

The undeniable roles of benzodiazepine-based drug in the treatment of diversities of infections cannot be over-emphasized as many commercially marketed drugs possess benzodiazepine as their core templates as shown in Table 2. Some of these drugs are chlordiazepoxide with sedative and anxiolytic properties (Prommer, 2020); diazepam with inhibitory properties which facilitate the action of gamma amino butyric acid (GABA) (Chakraborty, Sharmin, Rony, Ahmad, & Sohrab, 2018); clonazepam with anti-epileptic properties (Panahi et al., 2014); alprazolam with antidepressant activities (Rao, Ahmad, Madni, Ahmad, & Shahzad, 2020); lorazepam with anti-anxiety efficacy (Mercier et al., 2022); midazolam (marketed under the brand name Versed) possessed amnesic properties (Taghizadeh, Malakpour, & Javidan, 2019); delorazepam with muscle relaxant endowment (Magalhães et al., 2012) and oxazepam with anxiolytic and anticonvulsant properties (Chan, 2019; Varenne et al., 2022). The structures and therapeutic applications of the benzodiazepine-based drugs are as presented in Table 2. Benzodiazepines are used by Psychiatrists to treat nervousness, sleeping syndromes and liquor withdrawal and this afford relief at low doses when likened with barbiturates. They show fewer negative effects when likened with pyrimidinetrione motifs and dicarbamates (Pagel & Parnes, 2001; Shaikh et al., 2020). The long-term uses of benzodiazepines are prohibited by Food and Drug Administration but are rather permitted for short-range use base on the conditions (Casher, Botswick, & Yasugi, 2012).

3.1. Anxiolytics activity

Benzodiazepines are used by Psychiatrists to treat numerous anxieties for short-term administrations and provide relief at low doses. They are safer than...
both 5-ethyl-5-phenyl-2,4,6(1H,3H,5H)-pyrimidinetrione and dicarbamates (Pagel & Parnes, 2001). They are also applicable in the treatment or cure for nervousness disorders, and general unease syndromes, as additions for treatment of neurotic-driven condition (Griffin, Kaye, Bueno, & Kaye, 2013; Guina & Merrill, 2018). Examples of benzodiazepines used for relieving anxiolytics disorders are chlordiazepoxide, alprazolam, clonazepam, and lorazepam (Casher et al., 2012). The structures of these commercially marketed drugs for treatment of anxiolytics disorders are as given below (Figure 3).

### 3.2. Hypnotic activity

Benzodiazepines alter the rate of sleeping due to its ability to increase sleeping period by reducing the interval to fall asleep and the rate of arousals (Roehrs & Roth, 2010). Benzodiazepines reduce the deep sleep stages and increase the light-sleep phases. This decreases the most vital phase of sleep and affects sleep worth (Roehrs & Roth, 2010). Examples of commercially available benzodiazepines hypnotic drugs are temazepam, quazepam, flurazepam, and estazolam (Figure 4).

### 3.3. Muscle relaxant activity

Benzodiazepines such as alprazolam, diazepam, and lorazepam can be used as body relaxants decreasing the manner of muscle spasm and prevent increased muscle tone. The relaxant features are facilitated via α2-comprising receptors in the central nervous system. They could also inhibit the ache of the cerebral palsy caused by other central nervous system pathologies (Griffin et al., 2013). Recently, administration of vehicle of diazepam, indicated that the unilateral exodontia (AO group) promoted an increase of oxidative fibers in the contralateral side (Nascimento et al., 2020) (Figure 5).

### 3.4. Antiproliferative activity

According to Sharp et al. (2017), benzodiazepine motif exhibited viable antiproliferative action in number of definite leukemia and down regulation of gene that has the potential to cause cancer. The mixes showed production of major osteosarcoma cell varieties, indicating the viability of 1,2,3-triazolo-benzodiazepine products in tumor studies (Sharp et al., 2017). Benzodiazepine motif exhibited excellent antiproliferative activity with highly potent anticancer activity against five cancer cell lines at nanomolar concentration ranging from 5.83-12.60 nM (Pang et al., 2019). In addition, compound showed very good antiproliferative activity with IC50 of 2.50 μM (Lisowski et al., 2002) (Figure 6).

### 3.5. Anticonvulsant activity

Shao et al. (2018) reported that compound exhibited very good anticonvulsant activity via maximal electroshock test and enhanced antiepileptic activity with pentylenetetrazol test (ED50 value of 36.5 mg/kg, ED50 of 68.2 mg/kg) (Shao et al., 2018). Compounds and possessed methylated lactamic group which might have brought about higher efficacy which was even better than that diazepam, which was the standard drug used (Gaponov et al., 2016) (Figure 7).

### 3.6. Antibacterial activity

Verma et al. (2019) reported the design and formation of novel polyfunctionalized benzodiazepine derivatives. The in vitro activities of the formed compounds were carried out and compounds and showed excellent antibacterial activities against S. aureus and B. subtilis at MIC of 1.70 and 2.30 μM respectively. Whereas, compound also showed good activity against E. coli at MIC of 1.98 μM (Verma et al., 2019). A series of pyrazole bearing benzodiazepine designed by simple and cheap precursor were investigated for their binding potential against critical microbial target DNA gyrase. The experimental validation showed to have good activities against bacterial strain used (Desai, Joshi, & Khedkar, 2020) (Figure 8).
Table 2. Pharmacological properties of benzodiazepine-based drugs and their applications.

| S/N | Benzodiazepine-based drugs | Pharmacological properties | Applications |
|-----|-----------------------------|----------------------------|--------------|
| 1   | Clorazepate                  | Decreases abnormal electrical activity in the brain, treatment of anxiety, alcohol withdrawal use (Prommer, 2020). |
| 2   | Diazepam (Valium)            | Facilitate the action of gamma aminobutyric acid (GABA), treatment of anxiety, sedative, muscle-relaxant, treatment of convulsion and amnestic (Chakraborty et al., 2018, Iwao, Inoue, Hayashi, Yuasa, & Watanabe, 2004). |
| 3   | Clonazepam (Klonopin)        | Facilitate GABAergic transmission in the brain, treatment of panic disorders (Panahi et al., 2014; Mercier et al., 2022) |
| 4   | Alprazolam (Xanax)           | It acts on the brain and nerves by enhancing the effects of certain natural chemical in the body (Rao et al., 2020) |
| 5   | Lorazepam (Ativan)           | It acts on the brain and nerves by enhancing the effects of certain natural chemical in the body (Mercier et al., 2022) |

(continued)
3.7. Anticancer activity

Chen et al. (2014) described the establishment of benzodiazepines derivatives as new possible chemotherapeutic agents. Benzodiazepine bearing carboxamide, 39 exhibited worthy chemotherapeutic activities against human (lung, breast, colon, cervical and lewis lung) carcinoma (Chen et al., 2014). Compound 40 also exhibited very good antiproliferative activity with excellent inhibitory features (Misra et al., 2020). Midazolam, 41 inhibited cancer cell proliferation both in epithelial and mesenchymal types at IC_{50} of 5 μM (Lu et al., 2021) (Figure 9).

3.8. Antidepressant activity

Benzodiazepines play a significant role towards the central nervous system by virtue of their selective binding in selective protein regions known as GABA-A receptors which are found in the brain (Anderson, 2022). In the treatment of depression, benzodiazepine
is added to the antidepressant treatment to ease the anxiety and sleeplessness which go in tandem with depression (Bushnell, Stürmer, Gaynes, Pate, & Miller, 2017). Sharma et al. (2017) reported phenolic linked compound 42 and chlorophenyl-linked compound 43 as viable antidepressant when evaluated via behavioral despair test in mice. Clorazepate, 44 is an extensive-stand-in antidepressant benzodiazepine drug with eradication half-life greater than 24 h (Batíle, Lizano, Viñas, & Pujol, 2018). However, it is potentially risky to elderly people or patients with metabolism diseases (Figure 10).

3.9. Anti-tubercular activity
Anil et al. (2019) documented the preparation of diverse benzodiazepines derivatives and their in vitro
evaluation against *M. tuberculosis* and their potency showed compounds 45 and 46 have very good anti-tuberculosis potentials at MIC of 1.55 and 2.87 mg/mL (Anil et al., 2019). In addition, compound 47 also showed viable ant-tubercular with MIC 1.6 mg/mL (Kumar et al., 2018). Ongone et al. (2019) reported the formation of alkylated-benzodiazepin-2-one derivatives. The *in vitro* test carried out showed that compounds 48 and 49 have viable anti-inflammatory activity. Bhat and Kumar (2016) showed 50 as a good anti-inflammatory agent (Figure 11).

### 3.10. Anti-fungal activity

Caiana et al. (2021) documented the preparation of some benzodiazepine motifs and *therein vitro* evaluation against fungal strain species of genus *Sporothrix* and their potency showed compound 51 has a very good anti-fungal potentials (MIC 45-89 μg/mL and MFC 89-179 μg/mL) when compared to the standard Fluconazole (MIC 52-209 μg/mL and MFC 104-418 μg/mL) (Caiana et al., 2021). In addition, compound 52 also showed viable ant-fungal potential when compared with standard drug Itraconazole (Paulussen et al., 2014) (Figure 12).

#### 3.10.1. GABA receptor agonistic nature of Benzodiazepines (BDZs)

In recent times, studies have focused attention around α5-containing GABA<sub>A</sub> receptors. Even though, they have scarce occurrence of less than 5% of the total receptors and their circulation limited to hippocampus (Rudolph & Mohler, 2014), crystal structure of a human GABA<sub>A</sub> receptor reported has substantiated the crucial role of this phenomenon (Miller &
Figure 11. Selected benzodiazepine motifs with ant tubercular properties.

Figure 12. Selected benzodiazepine motifs with antifungal properties.

Figure 13. The diagram of the GABA<sub>A</sub> receptor and it's binding with benzodiazepines (Soyka, 2017).
Aricescu, 2014). Benzodiazepine is one of the most important drugs in the market and under clinical trial phase with laudable efficacy for the allosteric modulation of GABA<sub>A</sub> receptor (Sieghart, 2015). It was statistically established in 2008 that above 5% of adult population in United State used benzodiazepine drugs for the treatment of insomnia, anxiety and presurgical preparatory condition (Olfson, King, & Schoenbaum, 2015). There are also several isoforms of each subunit exploiting a process called alternative splicing: six alpha subtypes (α1,2,3,4,5,6), four-beta (β1,2,3,4), three gammas (γ1,2,3), and one delta (δ). The binding affinity of (R)- and (S)-forms of three chemotypes (diazepam, imidazobenzodiazepine, and triazolam) against GABA<sub>A</sub> receptors was investigated using radioligand displacement as well as electrophysiology (Elgarf et al., 2018). There are two GABA binding sites in the receptor and a single binding site for the BDZs which is located in the coupling (interphase) between an α subunit and a β subunit (Soyka, 2017) (Figure 13).

4. Conclusion

In summary, benzodiazepine is a highly endowed heterocyclic scaffold with essential features for excellent performance in therapeutic medicine. This review unveiled and garnered valuable information on chemistry and pharmacological properties of benzodiazepine motifs in therapeutic medicine. Their commonest synthetic approach is the one that involved the utilization of o-phenylenediamine as excellent precursor that undergoes cyclo-condensation reaction with diverse ketones under the influence of catalysis, glycerol recyclable solvent approach, conventional thermal cyclisation, microwave-assisted approach in solvent-free condition, environmentally friendly approach etc. In the last decade, interest on benzodiazepines has increased sporadically due to their fast-growing research applications in the design of drugs for the treatment of diverse infectious diseases. Hence, there is need for continuous studies to pinpoint targeted compounds of this class as hit-to-leading biomolecules and their lucrative privilege in fighting diseases. Therefore, the explorative study of diversities of scientific efforts on chemistry and pharmacological propensity of benzodiazepines derivatives highlighted herein might pave ways for the discovery of novel biomolecules with potential for human utilization through drug commercialization.

Authors’ contributions

OOA designed the work and partook in the original draft. OOT wrote the chemistry and part of pharmacological aspect, OSS supervised the work and partook in the conceptualization, APP wrote the pharmacological aspect. The original draft and the entire review were written through contributions of all authors. All authors have given approval to the final version of the review.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

All authors gratefully acknowledged Covenant University for the support for this work. OOA sincerely thank Royal Society of Chemistry for RSC Research Fund Grant with the Grant No: R21-2456856027.

ORCID

Olayinka O. Ajani @ http://orcid.org/0000-0002-3422-3478

References

Ajanl, O. O., Akande, M. M., October, N., Siyanbola, T. O., Aderohunmu, D. V., Akinsiku, A. A., & Olorunshola, S. J. (2019). Microwave assisted synthesis, characterization and investigation of antibacterial activity of 3-(5-(substituted-phenyl)-4,5-dihydro-1H-pyrazol-3-yl)-2H-chromen-2-one derivatives. Arab Journal of Basic and Applied Sciences, 26(1), 362–374. doi:10.1080/25765299.2019.1632141

Anderson, L. A. (2022). Benzodiazepines: Overview and use. Retrieved 2022, March 25, from https://www.drugs.com/article/benzodiazepines.html.

Anil, S. M., Shobith, R., Kiran, K. R., Swaroop, T. R., Mallesha, N., & Sadashiva, M. P. (2019). Facile synthesis of 1,4-benzodiazepine-2,5-diones and quinazolinones from amino acids as anti-tubercular agents. New Journal of Chemistry, 43(1), 182–187. doi:10.1039/C8NJ04936J

Arora, N., Dhiman, P., Kumar, S., Singh, G., & Monga, V. (2020). Recent advances in synthesis and medicinal chemistry of benzodiazepines. Bioorganic Chemistry, 97, 103668. doi:10.1016/j.bioorg.2019.103668

Avalone, R., Cosenza, F., Farina, F., Baraldi, C., & Baraldi, M. (2002). Extraction and purification from Ceratonia siliqua of compounds acting on central and peripheral benzodiazepine receptors. Fitoterapia, 73(5), 390–396. doi:10.1016/S0367-326X(02)001156

Azmí, S. N. H., Al-Fazari, A., Al-Badael, M., & Al-Mahrazi, R. (2015). Utility of Eosin Y as a complexing reagent for the determination of citalopram hydrobromide in commercial dosage forms by fluorescence spectrophotometry. Determination of citalopram hydrobromide. Luminescence: The Journal of Biological and Chemical Luminescence, 30(8), 1352–1359. doi:10.1002/bio.2905

Azmí, S. N. H., Al-Hadhrami, S. S. K., Al-Marhoubi, B. M. R., Al-Sulaimi, S. S. S., & Al-Shamoosi, Z. D. S. (2017). Development and validation of fluorescence spectrophotometric method: Quantitation of chlorohenimine-maléate in pharmaceutical formulation. Journal of
Molecular Liquids, 243, 750–760. doi:10.1016/j.molliq.2017.08.081
Azmi, S. N. H., Al-Masroui, Z. N., Al-Lamki, I. R., Al-Jabri, A. K., Rahman, N., Nasir, M., … Alam, M. (2022). Development and validation of spectrophotometric method for determination of imipramine hydrochloride in tablets (solid materials). Journal of King Saud University – Science, 34(2), 101823. doi:10.1016/j.jksus.2022.101823
Azmi, S. N. H., Iqbal, B., Al-Humaimi, S. M. H., Al-Salmani, I. R. S., Al-Ghafari, N. A. S., & Rahman, N. (2013). Quantitative analysis of cefixime via complexation with palladium(II) in pharmaceutical formulations by spectrophotometry. Journal of Pharmaceutical Analysis, 3(4), 248–256.
Azmi, S. N. H., Iqbal, B., Jabooob, M. A. M., Al Shahari, W. A. S., & Rahman, N. (2009). Spectrophotometric determination of piroxicam via chelation with Fe(III) in commercial dosage forms. Journal of the Chinese Chemical Society, 56(6), 1083–1091. 2009. doi:10.1007/jccs.200900157
Balaban, A. T., Onciu, D. C., & Katritzky, A. R. (2004). Catalytic asymmetry as a cornerstone of heterocyclic chemistry. Chemical Reviews, 104(5), 2777–2812.
Batile, E., Lízano, E., Viñas, M., & Pujol, M. D. (2018). 1,4-Benzodiazepines and new derivatives: Description, analysis, and organic synthesis. In J. Vasková, & L. Vaško (Eds.), Medicinal Chemistry, IntechOpen Limited, UK.doi: 10.5772/intechopen.79879
Bhat, K. I., & Kumar, A. (2016). Synthesis and anti-inflammatory activity of some novel 1,5-benzodiazepine derivatives. Asian Journal of Pharmaceutical and Clinical Research, 9(4), 63–66.
Buhl, T., Rosmarin, D., Serra-Baldrich, E., Fernandez-Peñas, P., Igarashi, A., Konstantinou, M. P., … Casillas, M. (2021). Itch and sleep improvements with baricitinib in patients with atopic dermatitis: A post hoc analysis of 3 phase 3 studies. Dermatology and Therapy, 11(3), 971–982. doi:10.1007/s13555-021-00534-8
Burman, R. J, Selfe, J. S., Lee, J. H., van den Berg, M., Calin, A., Codadu, N. K., … Raimondo, J. V. (2019). Excitatory GABAergic signalling is associated with benzodiazepine resistance in status epilepticus. Brain: A Journal of Neurology, 142(11), 3482–3501. doi:10.1093/brain/awz283
Bushnell, G. A., Stürmer, T., Gaynes, B. N., Pate, V., & Miller, M. (2017). Simultaneous antidepressant and benzodiazepine new use and subsequent long-term benzodiazepine use in adults with depression, United States, 2001-2014. JAMA Psychiatry, 74(7), 747–755. doi:10.1001/jamapsychiatry.2017.1273
Cacchi, S., Fabrizi, G., Goggianni, A., & Iazzetti, A. (2016). Construction of the 1,5-benzodiazepine skeleton from o-phenylenediamine and propargylalcohols via a domino gold catalyzedhydroxyamination/cyclization process. Organic Letters, 18(15), 3511–3513. doi:10.1021/acs.orglett.6b01720
Calana, E. C., de Veras, B. O., de Souza, A. L., & Queiroz, N. (2021). Synthesis of hydroxybenzodiazepines with potential antioxidant and antifungal action. Journal of the Brazilian Chemical Society, 32(3), 455. doi:10.21577/jbchs0103-5053.202000217
Calvo, A. M., Prado, M. T. O., Dionisio, T. J., Marques, M. P., Brozoski, D. T., Lanchote, V. L., … Santos, C. F. (2016). Effective method for the detection of piroxicam in human plasma using HPLC. Brazilian Oral Research, 30(1), 1–6. doi:10.1590/1807-3107BOR-2016.vol30.0058
Cashner, M. L., Botswick, J. R., & Yasugi, S. (2012). Benzodiazepines: A versatile clinical tool; evidence supports their use for alcohol withdrawal, insomnia, anxiety disorders, and other conditions. Current Psychiatry, 11(4), 54–64.
Chakraborty, S., Sharmin, S., Rony, S. R., Ahmad, S. A. I., & Sohrab, M. (2018). Stability-indicating UV/VIS spectrophotometric method for diazepam development and validation. Indian Journal of Pharmaceutical Sciences, 80(2), 366–373. doi:10.4172/pharmaceutical-sciences.1000366
Chan, J. M. (2019). Drug metabolism and pharmacogenetics. In Pharmacology and physiology for anesthesia (2nd ed., pp 70–90). Elsevier Publishers, Nederland. doi:10.1016/B978-0-323-48110-6.00004-1
Chen, Y., Le, V., Xu, X., Shao, X., Liu, J., & Li, Z. (2014). Discovery of novel 1,5-benzodiazepine-2,4-dione derivatives as potential anticancer agents. Bioorganic & Medicinal Chemistry Letters, 24(16), 3948–3951. doi:10.1016/j.bmcl.2014.06.041
Chen, Y., Liu, X., Shi, W., Zheng, S., Wang, D., & He, L. (2020). One pot synthesis of seven-membered heterocyclic derivatives of diazepines involving copper catalyzed rearrangement cascade allyl-amination. The Journal of Organic Chemistry, 85(8), 5146–5157. doi: 10.1021/acs.joc.9b02710
Christodoulou, M. S., Beccalli, E. M., & Giofré, S. (2020). Palladium-catalyzed benzodiazepines synthesis. Catalysts, 10(6), 634. doi:10.3390/catal10060634
Danta, M. C., De Oliveira, F. S., Bandeira, S. M., Batista, J. S., Silva, C. D., Jr, Alves, P. B., … Marchioro, M. C. (2004). Central nervous system effects of the crude extract of Erythrina velutina on rodents. Journal of Ethnopharmacology, 94(1), 129–133. doi:10.1016/j.jep.2004.05.007
Da, Silva, A. V., Meneghetti, S. M. P., & Meneghetti, M. R. (2021). Benzodiazepines: Drugs with chemical skeletons suitable for the preparation of metallacalixes with potential pharmacological activity. Molecules, 26(9), 2796. doi:10.3390/molecules26092796
Desai, N. C., Joshi, S. B., & Khedkar, V. M. (2020). Synthesis, antimicrobial activity and molecular docking of pyrazole bearing the benzodiazepine moiety. Analytical Chemistry Letters, 10(3), 307–320. doi:10.1080/22297928.2020.1785325
Dubovsky, S. L., & Marshall, D. (2022). Benzodiazepines remain important therapeutic options in psychiatric practice. Psychotherapy and Psychosomatics, 91(5), 307–328. doi:10.1159/000524400
Dzierzinski, F., Coppin, A., Mortuaire, M., Dewailly, E., Slomiany, C., Ameisen, J. C., … Tomavo, S. (2002). Ligands of the peripheral benzodiazepine receptor are potent inhibitors of Plasmodium falciparum and Toxoplasma gondii in vitro. Antimicrobial Agents and Chemotherapy, 46(10), 3197–3207. doi:10.1128/AAC.46.10.3197-3207.2002
Elgar, A. A., Siebert, D. C. B., Steudle, F., Draxler, A., Li, G., Huang, S., … Scholze, P. (2018). Different benzodiazepines bind with distinct binding modes to GABA_A receptors. ACS Chemical Biology, 13(8), 2033–2039. doi:10.1021/acschembio.8b00144
El-Sayad, H. I. H., Elkholy, W. M. E., & Hamed, W. A. E. (2017). Promising role of Carob (Ceratonia siliqua L.) phytochemical components against neurotoxicity induced by monosodium glutamate. Global Journal of Zoology, 2(1), 024–032. doi:10.17352/gjz.000008
Farhid, H., Khodkari, V., Nazeri, M. T., Javanbakht, S., & Shaabani, A. (2021). Multicomponent reaction of a domino gold catalysedhydroxyamination/cyclization process. ARAB JOURNAL OF BASIC AND APPLIED SCIENCES 303
Gaponov, A. A., Zlenko, E. T., Shishkina, S. V., Shishkin, O. V., Antypenko, O. M., Tretiakov, S. V., & Palchikov, V. A. (2016). Synthesis, spectroscopic characterization, X-ray structure, and in vivo neurotropic activity of new 1,5-benzodiazepin-2-ones. *Medicinal Chemistry Research*, 25(9), 1768–1780. doi:10.1007/s10004-016-1605-z

Griffin, C. E., Kaye, A. M., Bueno, F. R., & Kaye, A. D. (2013). Benzodiazepine pharmacology and central nervous system–mediated effects. *The Ochsner Journal*, 13, 214–223.

Guina, J., & Merril, B. (2018). Benzodiazepines I: Upping the care on downers: The evidence of risks, benefits and alternatives. *Journal of Clinical Medicine*, 7(2), 17. doi:10.3390/jcm7020017

Ilango, S. S., Remya, P. U., & Ponnuswamy, S. (2013). Synthesis and antimicrobial activity of novel 1,5-benzodiazepines. *ChemInform*, 44(no), 140. doi:10.1002/chin.201319191

Indalkar, K. S., Patil, M. S., & Chaturbhuj, G. U. (2017). An efficient, environmentally benign, and solvent-free protocol for the synthesis of 4-substituted 1,5-benzodiazepines catalyzed by reusable sulfated polyantrate. *Tetrahedron Letters*, 58(48), 4496–4502. doi:10.1016/j.tetlet.2017.

Insuasty, D., Robledo, S. M., Vélez, I. D., Cuervo, P., Insuasty, B., Quiroga, J., … Abonia, R. (2017). A Schmidt rearrangement-mediated synthesis of novel tetrahydro-benzo[1,4]diazepin-5-ones as potential anticancer and antiprotozoal agents. *European Journal of Medicinal Chemistry*, 141, 567–583. doi:10.1016/j.ejmech.2017.10.024

Isaeva, V. I., Timofeeva, M. N., Panchenko, V. N., Lukoyanov, I. A., Chernyshev, V. V., Kapustin, G. I., … Kustov, L. M. (2019). Design of novel catalysts for synthesis of 1,5-benzodiazepines from 1,2-phenylenediamine and ketones: NH₄-MIL-101(Al) as integrated structural catalyst for catalytic materials based on calix[4]arenes. *Journal of Catalysis*, 369, 60–71. doi:10.1016/j.jcat.2018.10.035

Iwao, T., Inoue, K., Hayashi, Y., Yuasa, H., & Watanabe, J. (2004). Absorption and metabolic elimination of diazepam from the perfused rat small intestine. *Drug Metabolism and Pharmacokinetics*, 19(6), 430–437. doi:10.2133/dmpk.19.430

Kim, S., Kim, S. A., Nam, G. H., Hong, Y., Kim, G. B., Choi, Y., … Kim, I. S. (2021). In situ immunogenic clearance induced by a combination of photodynamic therapy and rho-kinase inhibition sensitizes immune checkpoint blockade response to elicit systemic antitumor immunity against intraocular melanoma and its metastasis. *Journal for ImmunoTherapy of Cancer*, 9(1), e001481. doi:10.1136/jitc-2020-001481

Kumar, M. M. K., Mohan, T., Mai, G. K., Sangeeta, G. P. V., & Nagasree, K. P. (2018). Synthesis, characterization and biological evaluation of novel 1,4-benzodiazepine derivatives as potent anti-tubercular agents. *Journal of Young Pharmacists*, 10(3), 267–271. doi:10.5350/jyp.2018.10.60

Lian, G., Li, J., Liu, P., & Sun, P. (2019). Photoredox-catalyzed radical cascade reaction to synthesize fluorinated pyrrolo[1,2-d]benzodiazepine derivatives. *Journal of Organic Chemistry*, 84(14), 9322–9329. doi:10.1021/acs.joc.9b00937

Lisowski, V., Fabis, F., Pierre, A., Caingnard, D.-H., Renard, P., & Rault, S. (2002). Synthesis of new aromatic pyrrolo[2,1-c][1,4]benzodiazepines and pyrrolo[1,2-a]thieno[3,2-e][1,4]diazepines as anti-tumoral agents. *Journal of Enzyme Inhibition and Medicinal Chemistry*, 17(6), 403–407. doi:10.1080/1475636021000005712

Lu, H. L., Wu, K. C., Chen, C. W., Weng, H. K., Huang, B. M., Lin, T. Y., … Wang, Y. K. (2021). Anticancer effects of midazolam on lung and breast cancers by inhibiting cell proliferation and epithelial-mesenchymal transition. *Life*, 11(12), 1396. doi:10.3390/life11121396

Magalhães, E. J., Nascentes, N. S., Augusti, R., Queiroz, M. E., Silva, C. C., Jr, & Afonso, R. (2012). Fast determination of benzodiazepines in human urine via liquid-liquid extraction with low temperature partitioning and LC-HRMS. *American Journal of Analytical Chemistry*, 03(02), 118–124. doi:10.4236/ajac.2012.32017

Majid, S. A., Khanday, W. A., & Tomar, R. (2012). Synthesis of 1,5-benzodiazepine and its derivatives by condensation reaction using H-MCM-22 as catalyst. *Journal of Biomedicine & Biotechnology*, 2012, 510650. doi:10.1155/2012/510650

Mbomo, R. E. A., Omam, J. P. O., Kavaye, A. K., Kameni, S. J. N., & Bum, E. N. (2015). Anxiolytic (benzodiazepine-like) properties of Mimosas pudica in mice. *International Journal of Brain and Cognitive Science*, 4(3), 41–49. doi:10.5923/j.ibjcs.20150403.01

Mercier, B., Scala-Bertola, J., Pape, E., Kolodziej, A., Gibaja, V., Bisch, M., … Gambier, N. (2022). Online SPE UPLC-MS/MS method for the simultaneous determination of 33 psychoactive drugs from swab-collected human oral fluid samples. *Analytical and Bioanalytical Chemistry*, 414(14), 4203–4215. doi:10.1002/sab.202004739

Miller, P. S., & Aricescu, A. R. (2014). Crystal structure of a human GABAA receptor. *Nature*, 512(7514), 270–275. doi:10.1038/nature13293

Misra, A., Kishore, D., Verma, V. P., Dubey, S., Chander, S., Gupta, N., … Sharma, S. (2020). Synthesis biological evaluation and molecular docking of pyrimidine and quinazoline derivatives of 1,5-benzodiazepine as potential anticancer agents. *Journal of King Saud University – Science*, 32(2), 1486–1498. doi:10.1016/j.jsus.2019.12.002

Nascimento, G. C., Malzone, B. L., Iyomasa, D. M., Pereira, Y. C. L., Issa, J. P. M., Leite-Panissi, C. R. A., … Dias, F. J. (2020). Beneficial effects of benzodiazepine on masticatory muscle dysfunction induced by chronic stress and occlusal instability in an experimental animal study. *Scientific Reports*, 10(1), 8787. doi:10.1038/s41598-020-65524-w

Ogawa, Y., Takeshima, N., Hayasaka, Y., Tajika, A., Watanabe, N., Streiner, D., & Furukawa, T. A. (2019). Antidepressants plus benzodiazepines for adults with major depression. *The Cochrane Database of Systematic Reviews*, 6(6), CD001026. doi:10.1002/14651858.cd001026

Olfsen, M., King, M., & Schoenbaum, M. (2015). Benzodiazepine use in the United States. *JAMA Psychiatry*, 72(2), 136–142. doi:10.1001/jamapsychiatry.2014.1763

Ongone, T. N., Achour, R., El Ghoul, M., El Ouaisif, L., El Jenli, M., Chemlal, L., … Zellou, A. (2019). Analgesic and antioxidant activities of 4-phenyl-1,5-benzodiazepin-2-one and its long carbon chains derivatives. *Journal of Chemistry*, 2019, 1–7. doi:10.1155/2019/9043571

Pagel, J. F., & Parnes, B. L. (2001). Medications for the treatment of sleep disorders: An overview. *Primary Care Companion to the Journal of Clinical Psychology*, 3(3), 118–125. doi:10.4088/pcc.v03n03

Panahi, H. A., Mehramizi, A., Ghassiemi, S., & Moniri, E. (2014). Selective extraction of clonazepam from human plasma and urine samples by molecularly imprinted polymeric beads. *Journal of Separation Science*, 37(6), 691–695. doi:10.1002/jssc.201301144

Pang, Y., Lin, H., Ou, C., Cao, Y., An, B., Yan, J., & Li, X. (2019). Design, synthesis, and biological evaluation of novel benzodiazepine derivatives as anticancer agents through inhibition of tubulin polymerization in vitro and
in vivo. European Journal of Medicinal Chemistry, 182, 111670. doi:10.1016/j.ejmech.2019.111670

Pasha, M. A., & Jayashankara, V. P. (2006). An expeditious synthesis of 1,5-benzodiazepine derivatives catalysed by p-toluenesulfonic acid. Journal of Pharmacological and Toxicology, 116(5), 573–578. doi:10.3923/jpt.2006.573.578

Patro, G., Bhattamisra, S. K., & Mohanty, B. K. (2016). Effects of Mimosapudico L. leaves extract on anxiety, depression and memory. Avicenna Journal of Phytomedicine, 6(6), 696–710.

Paulussen, C., Wit, K., Boulet, G., Cos, P., Meerpoel, L., & Maes, L. (2014). Pyrrolidinyl[2-3x]benzodiazepines show potent in vitro antifungal activity and significant in vivo efficacy in a Microsporum canis dermatitis model in guinea pigs. The Journal of Antimicrobial Chemotherapy, 69(6), 1608–1610. doi:10.1093/jac/dku034

Pérez-Mayoral, E., & López-Peinado, A. J. (2021). Porous catalytic systems in the synthesis of bioactive heterocycles and related compounds. In Green synthetic approaches for biologically relevant heterocycles (2nd ed., Vol. 2, pp. 97–164), Elsevier Publishers, Netherland. doi:10.1016/B978-0-12-028792-5.00010-X

Prommer, E. (2020). Midazolam: An essential palliative care drug. Palliative Care and Social Practice, 2010.08.002

Qian, J., Liu, Y., Cui, J., & Xu, Z. (2012). Gold (1)-catalyzed synthesis of 1,5-benzodiazepine directly from o-phenylenediamines and alkynes. The Journal of Organic Chemistry, 77(9), 4484–4490. doi:10.1021/jo300543n

Radatz, C. A., Silva, R. B., Perin, G., Lenardao, E. J., Jacob, R. G., & Alves, D. (2011). Catalyst-free synthesis of benzoazepines and benzimidazoles using glycerol as recyclable solvent. Tetrahedron Letters, 52(32), 4132–4136. doi:10.1016/j.tetlet.2011.05.142

Rahman, N., Azmi, S., & Wu, H. F. (2006). The importance of impurity analysis in pharmaceut ical products: An integrated approach. Accreditation and Quality Assurance, 11(1-2), 69–74. doi:10.1007/s00769-006-0095-y

Rahman, N., Siddiqui, S., & Azmi, S. N. H. (2009). Spectrofluorimetric method for the determination of doxepin hydrochloride in commercial dosage forms. AAPS PharmSciTech, 10(4), 1381–1387. doi:10.1208/s12249-009-9341-z

Rao, H., Ahmad, S., Madni, A., Ahmad, I., & Shahzad, M. N. (2020). Single-step extraction for simultaneous quantifi-

Rashid, M. A., Ashraf, A., Rehman, S. S., Shahid, S. A., Mahmood, A., & Faruq, M. (2019). 1,4-Benzodiazepines synthesis, reaction and biological significance. Current Organic Synthetic, 16(5), 709–729. doi:10.20174/1570179416666190703113807

Ribeiro, M. D., Onusic, G. M., Poltronieri, S. C., & Viana, M. B. (2006). Effect of Erythrina velutina and Erythrina mulungu in rats submitted to animal model of anxiety and depression. Brazilian Journal of Medical and Biological Research – Revista Brasileira de Pesquisas Medicas e Biologicas, 39(2), 263–270. doi:10.1590/S0100-879X2006000200013

Roehrs, T., & Roth, T. (2010). Drug-related sleep stage changes: Functional significance and clinical relevance. Sleep Medicine Clinics, 5(4), 559–570. doi:10.1016/j.jsmc.2010.08.002

Rudolph, U., & Mohler, H. (2014). GABA_A receptor subtypes: Therapeutic potential in down syndrome, affective disorders, schizophrenia, and autism. Annual Review of Pharmacology and Toxicology, 54, 483–507. doi:10.1146/annurev-pharmtox-011613-135947

Sand, P., Kavvadias, D., Feineis, D., Riederer, P., Schreier, P., Kleinschnitz, M., … Beckmann, H. (2000). Naturally occurring benzodiazepines: Current status of research and clinical implication. European Archives of Psychiatry and Clinical Neuroscience, 250(4), 194–202. doi:10.1007/s004060070024

Shaikh, I. N., Baseer, M. A., Ahmed, D. B., Adil, S. F., Khan, M., & Alwarthan, A. (2020). Microwave assisted green synthesis of 1,5-benzodiazepines using Cu(II)-clay nano catalyst. Journal of King Saud University – Science, 32(1), 979–985. doi:10.1016/j.jksus.2019.08.001

Shao, Y.-P., Han, R.-B., H.-F., Wu, H.-F., F.-Y., & Piao, F.-Y. (2018). Synthesis and anticonvulsant activity of some novel 7-(benzylamino)-1H-benzo[b][1,4]diazepine-2,4(3H,5H)-dione derivatives. Medicinal Chemistry Research, 27(2), 642–652. doi:10.1007/s00044-017-2089-1

Sharma, R., Tilak, A., Thakur, R. N., Gangwar, S. S., & Sutar, R. C. (2017). Synthesis and pharmacological evaluation of substituted 1,5-benzodiazepine derivatives for its anti-depressant activity in experimental animals. World Journal of Pharmaceutical Research, 6(17), 925–931. doi:10.20959/wjpr201717-10410

Sharp, P. P., Garnier, J.-M., Hatfaludi, T., Xu, Z., Segal, D., Jarman, K. E., … Burns, C. J. (2017). Design, synthesis, and biological activity of 1,2,3-triazolobenzodiazepine BET bromodomain inhibitors. ACS Medicinal Chemistry Letters, 8(12), 1298–1303. doi:10.1021/acsmedchemlett.7b00389

Shobha, D., Chari, M. A., Selvan, S. T., Oveisi, H., Mano, A., Mukkanti, K., & Vinu, A. (2010). Room temperature synthesis of 1,5-benzodiazepine and its derivatives using cage type mesoporous aluminosilicate catalysts. Microporous and Mesoporous Materials, 129(1-2), 112–117. doi:10.1016/j.micromeso.2009.09.005

Shorter, E. (2005). Benzodiazepines: A historical dictionary of psychiatry (pp. 41–42). Oxford University Press, England.

Sieghart, W. (2015). Allosteric modulation of GABAA receptors via multiple drug-binding sites. Advances in Pharmacology (San Diego, Calif.), 72, 53–96. doi:10.1016/bs.apha.2014.10.002

Soyka, M. (2017). Treatment of benzodiazepine dependenc e. The New England Journal of Medicine, 376(12), 1147–1157. doi:10.1056/NEJMra1618132

Taghizadeh, M. J., malakpouri, G. r., & Javidan, A. (2019). Improved and scalable methods for the synthesis of midazolam drug and its analogues using isocyanide reagents. Journal of the Iranian Chemical Society, 16(4), 785–794. doi:10.1007/s13738-018-1555-0

Taha, A. M., & Rasheed, M. K. (2022). Synthesis and characterization of some 1,5-benzodiazepine derivatives from chalcones and their use as scavengers for some heavy metals in environmental systems. IOP Conference Series: Earth and Environmental Science, 961, 012094. doi:10.1088/1755-1315/961/1/012094

Teixeira-Silva, F., Santos, F. N., Sarasqueta, D. F. O., Alves, M. F. S., Neto, V. A., Paula, I. C. M., … Marchioro, M. (2008). Benzodiazepine-like effects of the alcohol extract from Erythrina velutina. leaves: Memory, anxiety, and epilepsy. Pharmaceutical Biology, 46(5), 321–328. doi:10.1080/13880200801887658

Trindade, P. A., Giglio, F. P., Colombini-Ishikiriama, B. L., Calvo, A. M., Modena, K. C., Ribeiro, D. A., … Santos, C. F. (2011). Comparison of oral versus sublingual
piroxicam during postoperative pain management after lower third molar extraction. *International Journal of Oral and Maxillofacial Surgery*, 40(3), 292–297. doi:10.1016/j.ijom.2010.10.026

Varenne, F., Kadhirvel, P., Bosman, P., Renault, L., Combès, A., & Pichon, V. (2022). Synthesis and characterization of molecularly imprinted polymers for the selective extraction of oxazepam from complex environmental and biological samples. *Analytical and Bioanalytical Chemistry*, 414(1), 451–463. doi:10.1007/s00216-021-03268-w

Varnonne, G. (2016). An update on the synthesis of pyrrolothiazepines. *Molecules (Basel, Switzerland)*, 21(2), 154. doi:10.3390/molecules21020154

Velasco-Rubio, A., Varela, J. A., & Saa, C. (2020). Recent advances in transition-metal-catalyzed oxidative annulation to benzazepines and benzodiazepines. *Advanced Synthesis & Catalysis*, 362(22), 4861–4875. doi:10.1002/adsc.202000808

Verma, D., Kumar, P., Narasimhan, B., Ramasamy, K., Mani, V., Mishra, R. K., & Majeed, A. B. A. (2019). Synthesis, antibacterial, anticancer and QSAR studies of 1-(4(substituted phenyl) 2-(substituted phenyl azomethyl)-benzo[b][1,4]diazepin-1-yl)-2-substituted phenyl aminoethanones. *Arabian Journal of Chemistry*, 12(8), 2882–2896. doi:10.1016/j.arabjc.2015.06.010

Verzijl, G. K. M., Hassfeld, J., Hassfeld, J., de Vries, A. H. M., & Lefort, L. (2020). Enantioselective synthesis of a 2,3-benzodiazepine intermediate of BET inhibitor BAY 1238097 via catalytic asymmetric hydrogenation. *Organic Process Research & Development*, 24(2), 255–260. doi:10.1021/acs.oprd.9b00519

Westland, J. L., & Dorman, F. L. (2013). QuEChERS extraction of benzodiazepines in biological matrices. *Journal of Pharmaceutical Analysis*, 3(6), 509–517. doi:10.1016/j.jpha.2013.04.004

Wicy, J. Y. (2013). The history of benzodiazepines. *Consultant Pharmacist*, 28(9), 538–548. doi:10.4140/TCP.n.2013.509

Wright, J. D., Cogan, J. C., Huang, Y., Tergas, A. I., St. Clair, C. M., Hou, J. Y., … Hershman, D. L. (2021). Association of new perioperative benzodiazepine use with persistent benzodiazepine use. *JAMA Network Open*, 4(6), e2112478. doi:10.1001/jamanetworkopen.2021.12478