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► **To cite this version:**

Maya Belhadj Mostefa, Amin Abedini, Laurence Voutquenne-Nazabadioko, Sophie C. Gangloff, Ahmed Kabouche, et al.. Abietane diterpenes from the cones of *Abies numidica* de Lannoy ex Carrière (Pinaceae) and in vitro evaluation of their antimicrobial properties. *Natural Product Research*, 2016, 31 (5), pp.568-571. 10.1080/14786419.2016.1190723 . hal-01996329

HAL Id: hal-01996329

<https://hal.univ-reims.fr/hal-01996329>

Submitted on 4 Oct 2021

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Abietane diterpenes from the cones of *Abies numidica* de Lannoy ex Carrière (Pinaceae) and *in vitro* evaluation of their antimicrobial properties

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Abstract

Eight known abietane diterpenes (**1-8**) were isolated for the first time from *Abies numidica* cones (Pinaceae). The structures of all compounds were established by means of 1D and 2D NMR, and UV spectral analyses. The hydromethanolic extract of *A. numidica* cones was tested for its antimicrobial activity against 17 Gram-positive and Gram-negative bacteria and against 5 yeasts by the use of liquid and solid medium and bioautography methods. The best antimicrobial activity was found against Gram-positive bacteria (MIC \leq 0.3mg/mL) *Bacillus subtilis*, *Enterococcus faecalis* ATCC 1034, *Staphylococcus aureus* 8325.4, *Staphylococcus aureus* CIP 53.154, *Micrococcus luteus* and *Listeria innocua* and against *Candida* yeasts. The determination of MIC's of isolated products showed a high activity of compounds **4** and **6** against *S. aureus*, *L. innocua* (MIC=62.5 μ g/mL) and *E. faecalis* (MIC=125 μ g/mL).

Keywords: *Abies numidica*; Pinaceae; abietane diterpenes; antimicrobial activity; solid medium; liquid medium; bioautography

1. Introduction

Abies is an important genus of the Pinaceae family. About 50 species are distributed in North Africa, Europe, North and Middle America and highlands of Asia (Zheng et al. 1978). Triterpenoids, steroids, flavonoids, phenols, fatty acids, and lignans have been previously reported (Yang et al. 2008). *Abies* species have exhibited several activities, including antitumour (Kim et al. 2004), anti-inflammatory, antihypertensive, anti-ulcerogenic (Singh et al. 2000), antibacterial (Vishnoi et al. 2007), antifungal (Aoyama et al. 1992), antitussive (Nayak et al. 2003), and central nervous system activities (Nayak et al. 2004). The cones of *Abies numidica* de Lannoy ex Carrière were used in folk medicine against cold, indigestion, stomachache, pulmonary, vascular and venereal diseases (Fujita et al. 1995). We report here, the phytochemical composition and the *in vivo* antimicrobial activity of the cones of *A. numidica*.

2. Result and discussion

Chromatography of the hydromethanolic extract of the cones of *A. numidica* afforded eight known compounds (1-8). They were unambiguously identified as: 7 α ,15-dihydroxydehydroabietic acid (1) (Prinz et al. 2002), 13 β -hydroxy-7-oxo-8(14)-abieten-18-oic acid (2) (Ohtsu et al. 2000), 15-hydroxy-7-oxo-dehydroabietic acid (3) (Matsumoto et al. 1988), 7 α -hydroxydehydroabietic acid (4) (Miguel Del Corral et al. 1994), 7-oxodehydroabietinol (5) (Tanaka et al. 1997), 7 α -hydroxy-15-methoxy-dehydroabietic acid (6) (Xian-Wen et al. 2010), 4-hydroxy-18-nor-8,11,13-abietatrien-7-one (7) (Lee et al. 1995) and 15-hydroxydehydroabietic acid (8) (Cheung et al. 1993) (figure 1) by comparison of their spectroscopic data with literature values.

The antimicrobial potential of the hydromethanolic extract of *A. numidica* cones was evaluated against 22 microorganisms including Gram-positive and Gram-negative bacteria as well as fungi by the MIC's evaluation in solid and liquid medium (Abedini et al. 2014; Acebey-Castellon et al. 2011). In solid media, the hydromethanolic extract was active at different degrees against 12 bacterial strains, particularly Gram-positive bacteria (Table 1). No antimicrobial activity was found against the Gram-negative bacteria strains, *Escherichia coli* CIP 54.127, *Enterobacter cloacae*, *Salmonella enterica*, *Klebsiella pneumoniae* and *Providencia stuartii*. The best antibacterial activity was obtained against six microorganisms namely *Bacillus subtilis*, *Enterococcus faecalis* ATCC 1034, *Staphylococcus aureus* 8325-4, *Staphylococcus aureus* CIP 53.154, *Micrococcus luteus* and *Listeria innocua* with MIC's values on agar below ≤ 0.3 mg/mL. The exact MIC values, in the liquid media, against these

six bacterial strains, reveal that the hydromethanolic extract have a strong activity against *Bacillus subtilis* (MIC=62.5 µg/mL) and a good antimicrobial activity against *E. faecalis*, *S. aureus* 8325-4, *S. aureus* CIP 53.154, *M. luteus* and *L. innocua* (MIC ≤250 µg/mL). This activity is very interesting for a crude extract (Toyang et al. 2012). To identify the compounds having an antibacterial activity in this extract, a bioautography assay (Abedini et al. 2013; Acebey-Castellon et al. 2011) was performed with the pure compounds (1-8) against *Staphylococcus aureus* CIP 53.154, one of the most sensitive microorganisms. Bioautography showed good to moderate activity for the five dehydroabietic acid derivatives (1, 3, 4, 6, 8) and low activity for compound 7 (Table 2). Therefore, these compounds were tested separately in liquid medium against *S. aureus* CIP53-154, *L. innocua* and *E. faecalis* ATCC 1034 (Table 2). Like bioautography, 4 and 6 showed the greatest activity against *S. aureus* and *L. innocua* (MIC=62.5 µg/ml) which is very close to MICs of antibiotics. They were less active against *E. faecalis* (MIC= 125µg/mL). Compound 1 showed a good activity against *L. innocua* (MIC=62.5µg/mL) and a moderate activity against *S. aureus* and *E. faecalis* (MIC= 125µg/mL). Compounds 3, 7 and 8 exhibited a moderate activity with MICs ranging between 125 and 500µg/mL. These results suggest that the presence of hydroxyl group in 7α position (1, 4, 6) may increase the antimicrobial activity of dehydroabietic acid derivatives compare to a ketone group (2, 3, 5, 7) as observed by Gouiric et al (2004). On the other hand, a carboxyl group in C-18 position in dehydroabietic acid derivatives seems to be more selective to the antibacterial activity.

The *in vitro* antifungal activity of the hydromethanolic extract was also tested against five yeasts and was active at 5mg/mL MIC's value against *Candida glabrata* and at 10mg/mL MIC's value against *Candida tropicalis*, *C. kefir* and *C. albicans* (Table 1).

3. Conclusions

Eight abietane diterpenes (1-8) were isolated for the first time from the cones of *Abies numidica*. The hydromethanolic extract was screened for their antibacterial and antifungal activities and demonstrated antibacterial activity against *Bacillus subtilis*, *Enterococcus faecalis*, *Staphylococcus aureus* 8325-4, *Staphylococcus aureus* CIP 53.154, *Micrococcus luteus* and *Listeria innocua* with MIC's ranging from 62.5 to 250 µg/mL.

Compounds 4 and 6 showed strong activity against *S. aureus* 8325-4 and *L. innocua* (MIC=62.5µg/mL), and compounds 1, 3, 7 and 8 a good antimicrobial activity. This work reveals that this plant can be used as potent sources of new antimicrobial agents against bacteria which are increasingly becoming resistant to traditional antibiotics.

Acknowledgment

The authors are grateful to ATRSS and DGRSDT (MESRS, Algeria), to university Mentouri Constantine-Algeria and to Groupe Isolement et Structure of the Institut de Chimie Moléculaire de Reims (ICMR), France for providing their financial support and facilities during the study. We also acknowledge Janick Madoux (Laboratory of Bacteriology, CHU Reims) for accessing to multiple inoculators material, Pr Jerome Depaquit (Laboratory of parasitology-Mycology) for providing yeasts and Chantal Grimplet for technical support.

Supplementary material

The experimental part of this article can be accessed on line at

References

- Abedini A, Roumy V, Mahieux S, Biabiany M, Standaert-Vitse A, Rivière C, Sahpaz S, Bailleul F, Neut C, Hennebelle T. 2013. Rosmarinic acid and its methyl ester as antimicrobial components of the hydromethanolic extract of *Hyptis atrorubens* Poit. (Lamiaceae). Evid Based Complement Alternat Med. 604536.
- Abedini A, Roumy V, Mahieux S, Gohari A, Farimani MM, Rivière C, Samaille J, Sahpaz S, Bailleul F, Neut C, Hennebelle T. 2014. Antimicrobial activity of selected Iranian medicinal plants against a broad spectrum of pathogenic and drug multiresistant microorganisms. Lett Appl Microbiol. 59(4): 412-421.
- Acebey-Castellon IL, Voutquenne-Nazabadioko L, Doan TMH, Roseau N, Boutaghane N, Muhammad D, Le Magrex-Debar E, Gangloff S, Litaudon M, Sevenet T, Hung NV, Lavaud C. 2011. Triterpenoids saponins from *Symplocos lancifolia*. J Nat Prod. 74:163-168.
- Aoyama M, Doi S.1992. Antifungal activities of wood extractives of todomatsu, *Abies sachalinensis* Masters against pathogenic fungi causing turfgrass diseases. Mokuza Gakkaishi. 38:101-105.
- Cheung HTA, Miyase T, Lenguyen MP, Smal MA. 1993. Further neutral acidic constituents of *Pinus massoniaca* resin. Tetrahedron. 9:7903-7915.
- Fujita T, Sezik E, Tabata M, Yesilada E, Honda G, Takeda Y, Tanaka T, Takaishi Y. 1995. Traditional medicine in Turkey. VII. Folk medicine in middle and west Black Sea regions. Economic Botany. 49:406- 422.

- Gouiric SC, Feresin G.E, Tapia AA, Rossomando PC, Schmeda-Hirschmann G, Bustos DA. 2004. $1\beta,7\beta$ -Dihydroxydehydroabietic acid, a new biotransformation product of dehydroabietic acid by *Aspergillus niger*. *World Journal of Microbiology & Biotechnology* 20: 281–284.
- Kim HJ, Choi EH, Lee IS. 2004. Two lanostane triterpenoids from *Abies koreana*. *Phytochemistry*. 65:2545-2549.
- Lee CK, Fang JM, Cheng YS.1995. Norditerpenes from *Juniperus chinensis*. *Phytochemistry*. 39:391-394.
- Matsumoto T, Imai S, Sunaoko Y, Yoshinari T. 1988. The conversion of (+)-dehydroabietic acid into steroidal. *Bull Chem Soc Jpn*.61:723-727.
- Miguel Del Corral JM, Gordaliza M, Salinero MA, San Feliciano A. 1994.¹³C-NMR data for abieta-8,11,13-triene diterpenoids. *Magn Reson Chem*. 32:774-781.
- Nayak SS, Ghosh AK, Srikanth K, Debnath B, Jha T. 2003. Antitussive activity of *Abies webbiana* Lindl. leaf extract against sulphur dioxide-induced cough reflex in mice. *Phytother Res*. 17:930- 932.
- Nayak SS, Ghosh AK, Debnath B, Vishnoi SP, Jha T. 2004. Synergistic effect of methanol extract of *Abies webbiana* leaves on sleeping time induced by standard sedatives in mice and anti-inflammatory activity of extracts in rats. *J Ethnopharmacol*. 93:397-402.
- Ohtsu H, Tanaka R, In Y, Matsunaga S, Tokuda H, Nishino H. 2000. New abietane diterpenoids from the cones of *Larix kaempferi*. *Can J Chem*. 78:31-40.
- Prinz S, Mullner U, Heilimann J, Winkelmann K, Sticher O, Haslinger E, Hufner A. 2002. Oxidation products of abietic acid and its methyl ester. *J Nat Prod*. 65:1530-1534
- Singh RK, Bhattacharya SK, Acharya SB. 2000. Pharmacological activity of *Abies pindrow*. *J Ethnopharmacol*.73:47- 51.
- Tanaka R, Ohtsu H, Matsunaga S. 1997. Abietane diterpene acids and other constituents from the leaves of *Larix kaempferi*. *Phytochemistry*. 46:1051-1057.
- Toyang NJ, Ateh EN, Keiser J, Vargas M, Bach H, Tane P, Sondengam LB, Davis H. 2012. Toxicity, antimicrobial and anthelmintic activities of *Vernonia guineensis* Benth. (Asteraceae) crude extracts. *J Ethnopharmacol*.144: 700–704.
- Vishnoi SP, Ghosh AK, Debnath B, Samanta S, Gayen S, Jha T. 2007. Antibacterial activity of *Abies webianna*. *Fitoterapia*.78:153- 155.
- Xian-Wen Y, Lin F, Su-Mei L, Xiao-Hua L, Yong-Li L, Liang W, Yun-Heng S, Jun-Mian T, Xi Z, Xin-Ru L, Ning W, Yonghong L, Wei-Dong Z. 2010. Isolation, structure and

bioactivities of abiesadines A-Y, 25 new diterpenes from *Abies georgei* Orr. *Bioorg. Med Chem.*18:744-754.

Yang XW, Li SM, Feng L, Shen YH, Tian JM, Liu XH, Zeng HW, Zhang C, Zhang WD. 2008. *Abies* anordines A-N: fourteen new norditerpenes from *Abies georgei*. *Tetrahedron.* 64:4354-4362

Zheng WJ, Fu LG. 1978. *Flora Reipublicae Popularis Sinicae* K, Ed. Z. Y. Wu. Science Press, Beijing, 55.