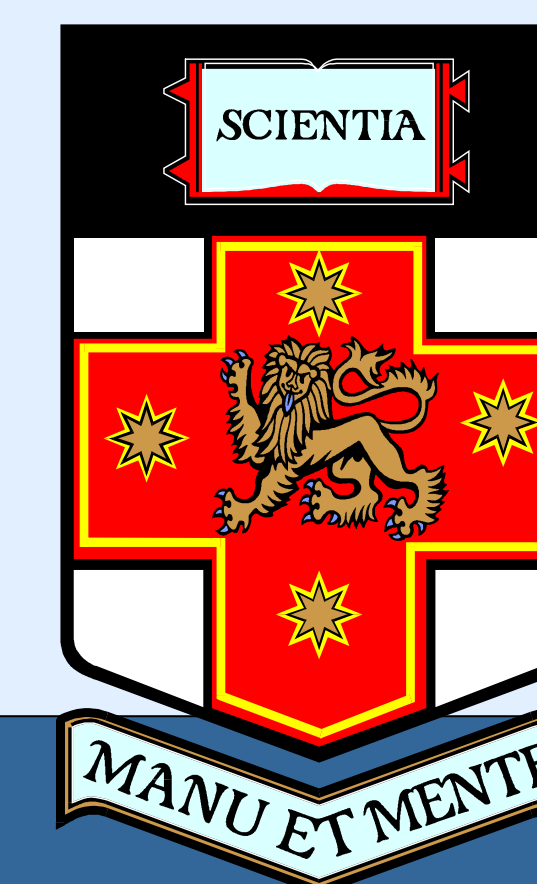


# Structural analysis of triterpenoid from genus *Pistacia* and their antimicrobial activity



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## Background

Isolated triterpenoids from acidic fractions of the tree trunk exudates from the genus *Pistacia* were isolated and structurally analysed. These isolates were divided into two major groups based on their skeleton; being lanosta, a tetracyclic triterpenoid compound, from which all steroids are derived, and oleanane, a natural triterpene found in flowering plants which are referred to collectively as oleanane triterpenes. The triterpenoids, lanosta, mimic certain steroidal-like compounds, particularly the well known antibiotic Fusidic acid. Selected isolates exhibited antibiotic properties and were tested in a protein inhibition assay to investigate their potential mode of action.

## Method

Column Chromatography, High Performance Liquid Chromatography and GC-Mass Spectrometry were used to isolate and characterised these chemical entities. The isolates were screened for anti-microbial activities and their Minimum Inhibitory Concentration (MIC) and kill kinetics were determined. The most bioactive components were structurally analysed and their mode of action/s were tested and compared with Fusidic acid by Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE).

## Results

The results of co-sedimentation of ribosome with Fusidic acid/ test compound was read by (SDS-PAGE). These results showed some similarity of lanosta type skeleton's mode of the action to mechanism of the action of Fusidic acid. However, the oleanane type structures did not exhibit the pattern observed with Fusidic acid and the lanosta type compounds. Notwithstanding, the negative results obtained for the oleanane type structures are very important from the perspective of structure and function analysis, as these compounds could appear to act by a mechanism distinct from steroidal antibiotics such as Fusidic acid. Potentially these compounds represent a new family of antimicrobial compounds.

## Results

Compound's No. (CN)	Isolated components subjected to SDS-PAGE	Lane No. on the Gel	Incubated at 0° for 20 min.
1	blank	2	Purified Ribosome (PR) (incubated with buffer + 1mM GTP)
2	fusidic acid	4,18-19	PR (incubated with 1mM CN2 + 1mM GTP) co-sedimentated with EF-G
3	moronic acid	3	PR (incubated with 1mM CN3 + 1mM GTP)
4	oleanonic acid	11	PR (incubated with 1mM CN4 + 1mM GTP)
5	ursonic acid	12	PR (incubated with 1mM CN5 + 1mM GTP)
6	oleanolic acid	13	PR (incubated with 1mM CN6 + 1mM GTP)
7	acidic Fractions (kurdia gum)	5	PR (incubated with 1mM CN7 + 1mM GTP)
8	isomasticadienonic acid	6	PR (incubated with 1mM CN8 + 1mM GTP) co-sedimentated with EF-G
9	3-epi-isomasticadienonic acid	7	PR (incubated with 1mM CN9 + 1mM GTP) co-sedimentated with EF-G
10	masticadienonic acid	8	PR (incubated with 1mM CN10 + 1mM GTP) co-sedimentated with EF-G
11	dihydromasticadienonic acid	9	PR (incubated with 1mM CN11 + 1mM GTP) co-sedimentated with EF-G
12	3-O-acetyl-3-epi(iso)masticadienonic acid	10	PR (incubated with 1mM CN12 + 1mM GTP) co-sedimentated with EF-G
13	masticadienonic acid	14	PR (incubated with 1mM CN13 + 1mM GTP) co-sedimentated with EF-G
14	dihydromasticadienonic acid	15	PR (incubated with 1mM CN14 + 1mM GTP) co-sedimentated with EF-G
15	3-acetoxy-3-epiisomasticadienonic acid	16	PR (incubated with 1mM CN15 + 1mM GTP) co-sedimentated with EF-G
16	3-acetoxy-3-epimasticadienonic acid	17	PR (incubated with 1mM CN16 + 1mM GTP) co-sedimentated with EF-G

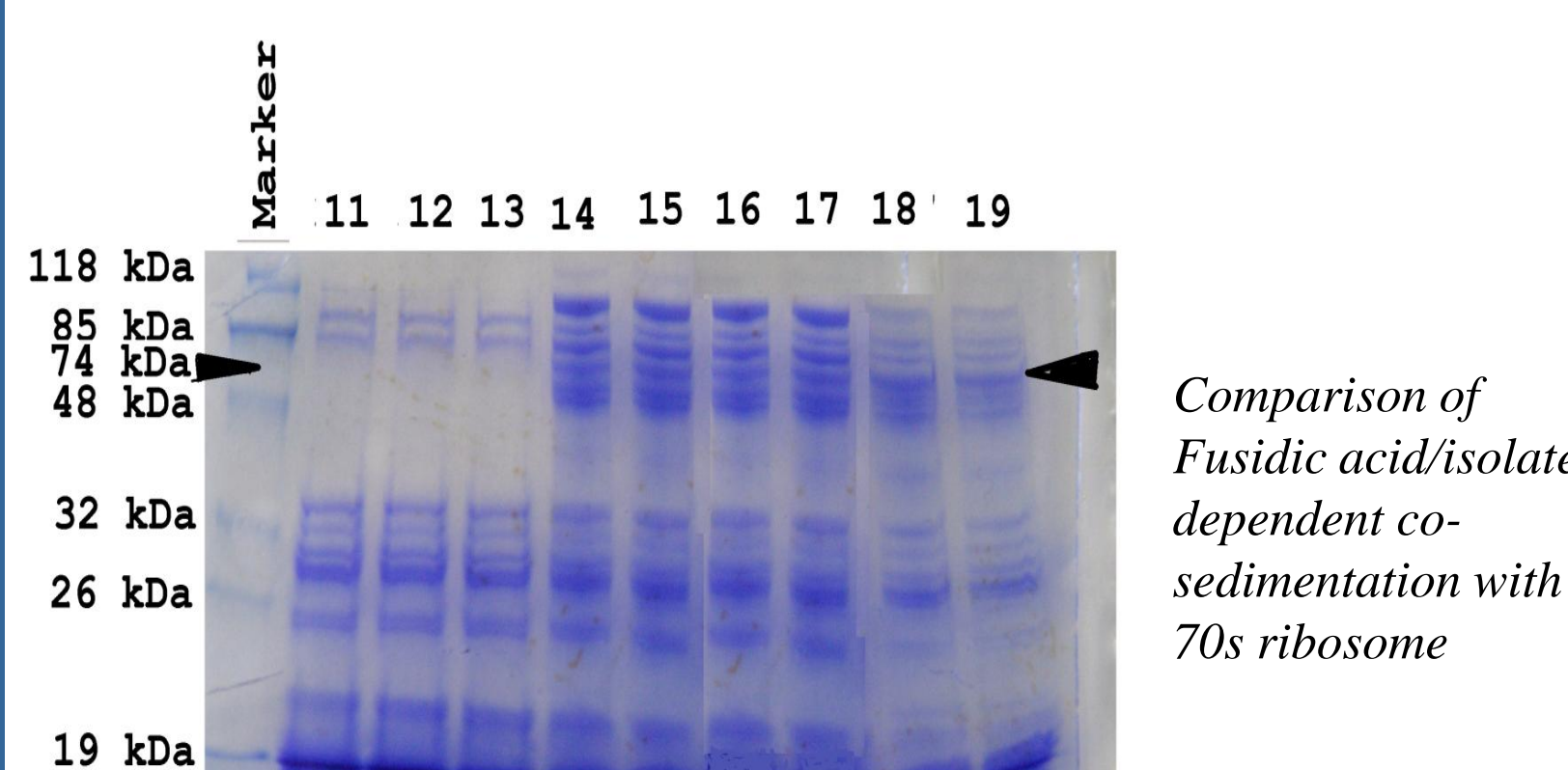
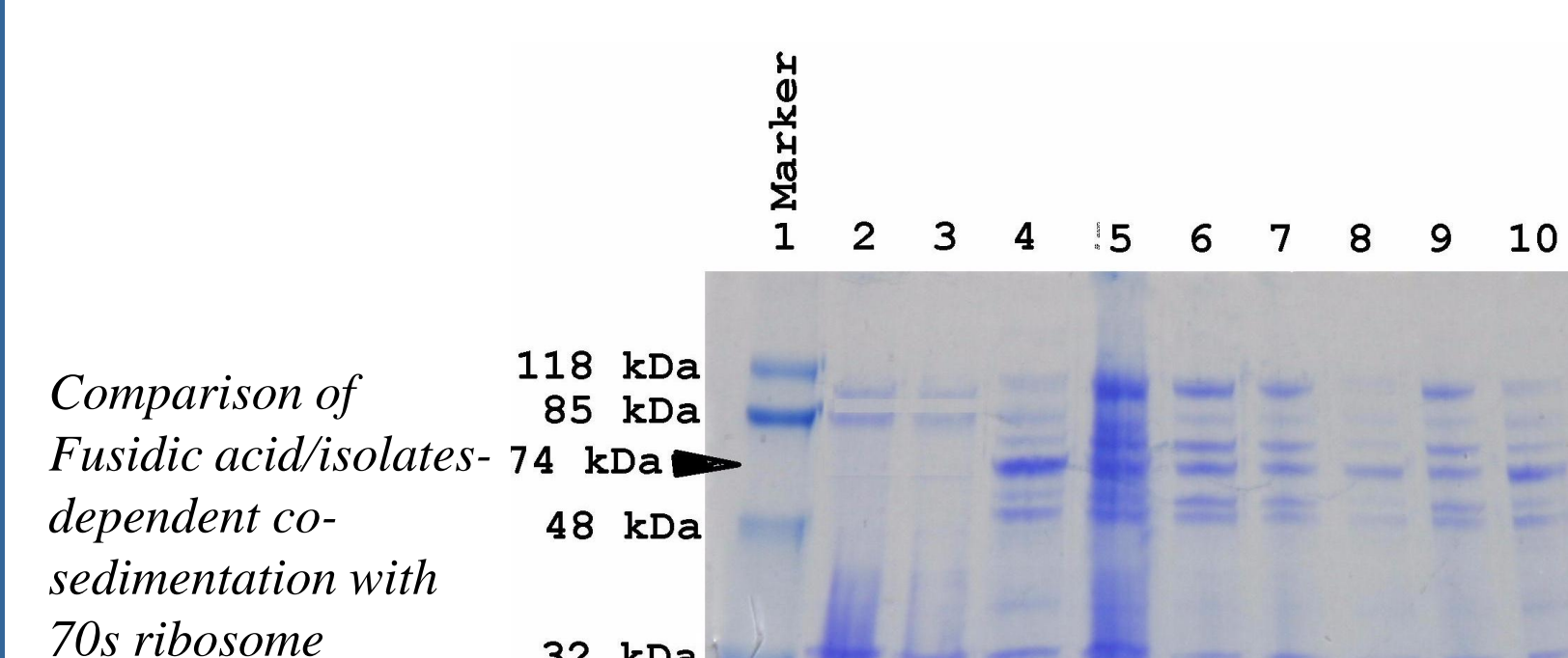
The summary of Fusidic acid/isolates subjected to SDS-PAGE electrophoresis

## Chemical Structures and MIC of the isolates

Structure I	MIC	Structure II	MIC	Structure III	MIC
	<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL		<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL		<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL
a: R=O, R1=COOMe Monsate	>100 100	a: R=O, R1=COOMe Oleanonate	>100 100	a: R=H, α-OH, R1=COOH Urosolic acid	100 50
b: R=O, R1=COOH Monsic acid	50 10	b: R=O, R1=COOH Oleanonic acid	50 10	b: R=H, α-OH, R1=COOH Urosolic acid (methyl ester)	>100 100
		c: R=H, β-OH, R1=COOMe Oleanolate	>100 100		
		d: R=O, R1=COOH Oleanolic acid	50 20		

Structure IV	MIC	Structure V	MIC	Structure VI	MIC
	<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL		<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL		<i>S. aureus</i> / <i>H. pylori</i> 26695 µg/mL
a: R=O, R1=COOMe Masticadienonate	>100 100	a: R=O, R1=COOMe Dihydromasticadienonate	>100 100	a: R=O, R1=COOMe Isomasticadienonate	>100 100
b: R=O, R1=COOH Masticadienonic acid	5 5	b: R=O, R1=COOH Dihydromasticadienonic acid	5 1	b: R=O, R1=COOH Isomasticadienonic acid	5 5
c: R=H, β-OH, R1=COOMe Masticadienolate	>100 100	c: R=H, β-OH, R1=COOMe Dihydromasticadienolate	>100 100	c: R=H, α-OH, R1=COOMe 3-epi-isomasticadienolate	>100 100
d: R=H, β-OH, R1=COOH Masticadienonic acid	10 5	d: R=H, β-OH, R1=COOH Dihydromasticadienonic acid	2 0.5	d: R=H, α-OH, R1=COOH 3-epi-isomasticadienonic acid	2 0.1
e: R=O, R1=COOMe 3-acetoxy-3-epimasticadienolate	>100 100	e: R=O, R1=COOMe 3-acetoxy-3-epimasticadienonic acid	>100 100	e: R=H, α-OH, R1=COOMe 3-O-acetyl-3-epi-isomasticadienolate	>100 100
f: R=O, R1=COOH 3-acetoxy-3-epimasticadienonic acid	5 0.5	f: R=O, R1=COOH 3-acetoxy-3-epimasticadienonic acid	5 0.5	g: R=H, α-OH, R1=COOMe 3-O-acetyl-3-epi-isomasticadienonic acid	>100 100
				h: R=H, α-OH, R1=COOH 3-O-acetyl-3-epimasticadienonic acid	0.05 0.01



## Conclusion

These chemical entities have produced promising data that could lead to the development of a novel class of antimicrobial agents that may have application in the treatment of infectious disease.

## Acknowledgements

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