Characterization of the new synthetic cannabinoid ‘Cumyl-PEGACLONE’ including first pharmacological data

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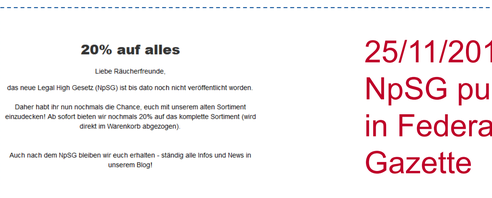
Introduction

Shortly after the German law on new psychoactive substances (NpSG) came into force on November 26th 2016, the first retailers of ‘herbal blends’ promoted new products not violating the German NpSG. We describe the identification and structural elucidation of one of the first synthetic cannabinoids not being covered by the NpSG.

Methods



25/11/2016
NpSG published in Federal Gazette

20/11/2016
Newsletter of legal high shop X: sale because of NpSG coming soon

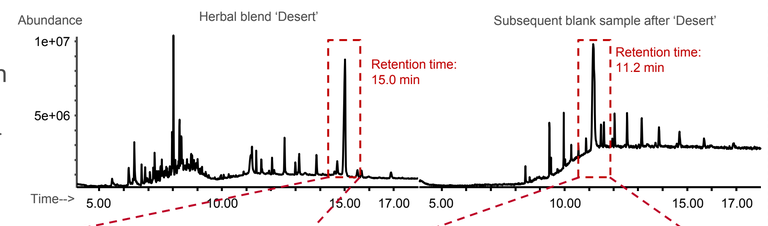
Newsletter of legal high shop X: no sale because of NpSG, but new products are on their way

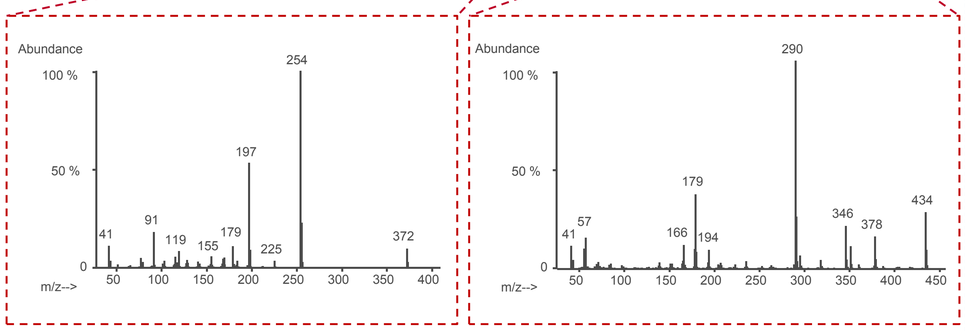
4/12/2016
Newsletter of legal high shop X: new products available

4/12/2016
Order of the available products

Extraction with ethanol

GC-MS

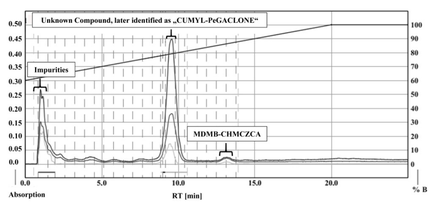




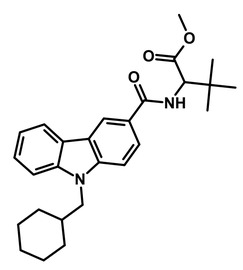
Library search (in-house library)

unknown

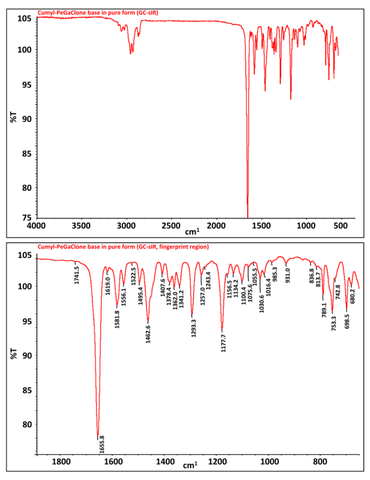
Isolation with flash chromatography



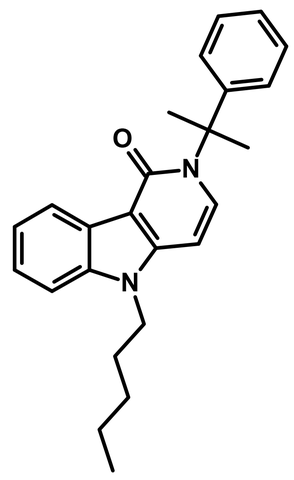
LC-ESI-qToF-MS
1D-/²D-NMR



Match with MDMB-CHMCZCA



Gas chromatography solid-state infrared analysis (GC-sIR)



$C_{25}H_{28}N_2O$
(measured accurate mass *m/z* 373.2271)
Suggested name: Cumyl-PEGACLONE

Measurement of binding affinities to human CB_1 and CB_2 receptors using Chinese hamster ovary cells and the CB agonist radioligand [^{3}H]-CP55,940 (details see [1])

Results and Discussion

The structure of the new synthetic cannbinoid was resolved as 5-pentyl-2-(2-phenylpropan-2-yl)-2,5-dihydro-1*H*-pyrido[4,3-*b*]indol-1-one by ^{1}H-,^{13}C-, HMBC-NMR and HRMS experiments This compound is structurally related to CUMYL-PICA, with the nitrogen atom of the amide function connected to the 2-position of the indole core via an ethene bridge, resulting in a tricyclic γ-carbolinone structure (see Fig. 1).

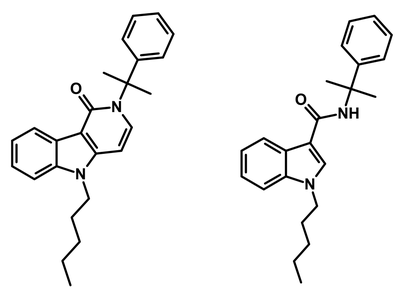


Fig 1:
Structural formula of Cumyl-PEGACLONE and Cumyl-PICA (also called ‘SGT-56’)

Table 1: Affinities at the human cannabinoid CB_1 and CB_2 receptors of the new substance and other substances (means ± SEM of 3 -4 separate experiments performed in duplicates)

	Human CB_1	Human CB_2
	K_i (nM)	K_i (nM)
Cumyl-PEGACLONE (FB-1)	1.37 ± 0.24	2.09 ± 0.33
JWH-019	8.13 ± 1.46 Aung *et al.* K_i = 9.80 nM [2]	6.58 ± 0.73 Aung *et al.* K_i = 5.55 nM [2]
Cumyl-PICA	3.27 ± 0.32	24.0 ± 8.8
EG018	7.17 ± 1.27	2.27 ± 0.38
EG2201	22.4 ± 12.8	4.36 ± 2.91

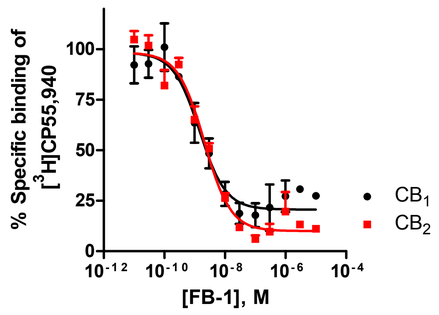


Fig 2:
Concentration-dependent inhibition of [^{3}H]CP55,940 binding by FB-1

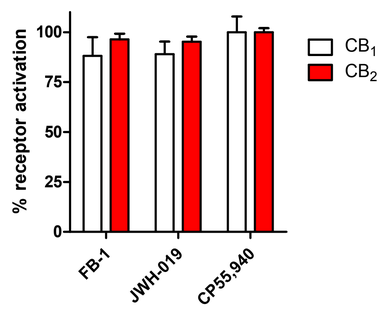


Fig 3:
cAMP accumulation assays for FB-1 and JWH-019 (concentration: 1 µM)

Cumyl-PEGACLONE is a strongly binding cannabinoid receptor ligand with K_i values in the low nanomolar range. It displays equal affinity for both cannabinoid receptors. Cumyl-PEGACLONE showed higher affinities to both receptors in direct comparison with Cumyl-PICA (see Table 1).

The CB receptor activation by Cumyl-PEGACLONE and JWH-019 was determined in cAMP accumulation assays. Receptor activation by an agonist results in a decrease in intracellular cAMP levels. Cells were preincubated with forskolin (10 µM) to elevate cAMP levels followed by the addition of the test compound (see Fig. 2). As shown in Fig. 3, Cumyl-PEGACLONE and JWH-019 both showed comparable effects on cAMP levels as the full agonist CP55,940 (concentration 1µM). Thus, all 3 compounds can be regarded as full CB_1 and CB_2 agonists.

Conclusion

The new compound bears a core structure circumventing the German NpSG as well as the generic definitions in other national laws. As a semi-systematic name for 2-cumyl-5-pentyl-gamma-carbolin-1-one Cumyl-PEGACLONE is suggested.

Acknowledgements

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References

[1] Hess, C.; Schoeder, C. T.; Pillaiyar, T.; Madea, B.; Müller, C. E. Pharmacological evaluation of synthetic cannabinoids identified as constituents of spice. *Forensic Toxicol.* **2016**, *34*, 329–343.

[2] Aung, M. M.; Griffin, G.; Huffman, J. W.; Wu, M.; Keel, C.; Yang, B.; Showalter, V. M.; Abood, M. E.; Martin, B. R. Influence of the N-1 alkyl chain length of cannabimimetic indoles upon CB_1 and CB_2 receptor binding. *Drug. Alcohol. Depend.* **2000**, *60*, 133–140.

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