Sinteza, karakterizacija i biološko djelovanje karbamidnih, semikarbazidnih, karbazidnih i esterskih derivata aminokiselina i nesteroidnih protuupalnih lijekova

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SVEUČILIŠTE U ZAGREBU
FARMACEUTSKO-BIOKEMIJSKI FAKULTET

IVANA PERKOVIĆ

SINTEZA, KARAKTERIZACIJA I BIOLOŠKO DJELOVANJE KARBAMIDNIH, SEMIKARBAZIDNIH, KARBAZIDNIH I ESTERSKIH DERIVATA AMINOKISELINA I NESTEROIDNIH PROTUUPALNIH LIJEKOVA

DOKTORSKI RAD

Zagreb, 2012.

UNIVERSITY OF ZAGREB
FACULTY OF PHARMACY AND BIOCHEMISTRY

IVANA PERKOVIĆ

SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF CARBAMIDE, SEMICARBAZIDE, CARBAZIDE AND ESTER DERIVATIVES OF AMINO ACIDS AND NONSTEROIDAL ANTIINFLAMMATORY DRUGS

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DOKTORSKI RAD

Mentor: Prof. dr. sc. Branka Zorc

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Rad je predan na ocjenu Fakultetskom vijeću Farmaceutsko-biokemijskog fakulteta Sveučilišta u Zagrebu radi stjecanja akademskog stupnja doktora znanosti iz područja biomedicine i zdravstva, polje farmacija, grana farmacija.

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SAŽETAK

U okviru ovog doktorskog rada, koji je nastavak istraživanja o mogućnostima korištenja benzotriazola u sintetskoj kemiji na Zavodu za Farmaceutsku kemiju Farmaceutsko-biokemijskog fakulteta u Zagrebu, sintetizirano je nekoliko serija spojeva. Polazni spoj u sintezi svih prekursora bio je klorid 1-benzotriazolkarboksilne kiseline (BtcCl) koji daje vrlo reaktivne derivate. BtcCl vrlo lako reagira s nukleofilima dajući cijeli niz reaktivnih spojeva iz kojih je do sada pripravljen velik broj organskih spojeva. Derivati aminokiselina ureidoamidi **4a**-**o** pripravljeni su reakcijom klorida *N*-(1-benzotriazol-karbonil)aminokiselina **3** i odgovarajućih aminoalkohola, a *N*- i *O*- supstituirane hidroksiuree **6a**-**l** iz amida *N*-(1-benzotriazolkarbonil)aminokiselina i odgovarajućih hidroksilamina. Derivati nesteroidnih protuupalnih lijekova (NSAID, ibuprofena, fenoprofena i ketoprofena) sintetizirani su iz NSAID hidrazida **11** koji su dobiveni aminolizom NSAID benzotriazolida **10** i hidrazina. Hidrazidi **11** su u reakciji s 1-karbamoilbenzotriazolima (aktivne uree, **7**) dali 1-acilsemikarbazidne derivate NSAID **13a**-**v**, a u reakciji s 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazidom (**9**) 1-acil-5-benziloksikarbamoilkarbazide **14a**-**c**. Reakcije u talini uz TEA bitno su skratile vrijeme reakcije. 4-Hidroksisemikarbazidni derivati NSAID **13w**-**y** i **14d**-**f** pripravljeni su katalitičkim hidrogeniranjem *O*-benzilnih derivata **13t**-**v** i **14a**-**c** uz Pd/C. Iz benzotriazolida ketoprofena (**10d**) i odgovarajućih ureidoamida (**4b**,**i**,**j**,**m**) pripravljeni su esteri s dvije molekule ketoprofena kovalentno povezane ureidoamidima u jednu molekulu („dvojni lijekovi“).

Svi sintetizirani spojevi karakterizirani su uobičajenim analitičkim postupcima i spektroskopskim metodama te biološki ispitani *in vitro* na citostatsko, antivirusno, antimikrobno i antioksidativno djelovanje. Nadalje, ispitana je inhibicija lipooksigenaze i lipidne perkosidacije. Najbolje antitumorsko djelovanje pokazao je spoj **6l**, antivirusno **13a**,**g**,**m**, a antiooksidativno spoj **9**. Najjači inhibitor lipooksigenaze bio je spoj **13s**, a lipidne peroksidacije **13c**,**l**,**t**. Uočeno je ponavljanje benzhidrilnog supstituenta u spojevima s istaknutom aktivnosti, što je dobar pokazatelj za daljnje usmjeravanje istraživanja.

Ključne riječi: aminokiselina/hidroksiurea/nesteroidni protuupalni lijek/ semikarbazid/ester/sinteza/biološko djelovanje

SUMMARY

The research described in the thesis is a continuation of the previous work on the use of benzotriazole in the synthetic chemistry which has been carring out at Department of Medicinal Chemistry, Faculty of Pharmacy and Biochemistry in Zagreb for many years. The thesis includes synthesis of several types of compounds. The initial compound for the synthesis of all the intermediates was benzotriazole carboxylic acid chloride (BtcCl), which upon the reaction with nucleophiles gives reactive products, useful in the preparation of various organic compounds. Amino acid derivatives ureidoamides **4a**-**o** were syntehsized by aminolysis of *N*-(1-benzotriazolecarbonyl)amino acid chlorides **3** with corresponding aminoalcohols and *N*- i *O*- substituted hydroxyureas **6a-l** were synthesizes from *N*-(1-benzotriazolecarbonyl)amino acid amides **5** and hydroxylamines. Nonsteroidal antiinflammatory drugs (NSAID, ibuprofen, fenoprofen and ketoprofen) derivatives were prepared from NSAID hydrazides **11** which were obtained by aminolysis of NSAID benzotriazolides **10** and hydrazine hydrate. The reaction of hydrazides **11** and 1-benzotriazole carboxylic acid amides **7** gave 1-acylsemicarbazides **13a**-**v**, while with 1-(1-benzotriazolecarbonyl)-4-benzyloxysemicarbazide (**9**) gave 1-acyl-5-benzyloxycarbamoyl-carbazides **14a**-**c**. Reactions in melted state in the presence of TEA significantly reduced the reaction time. 4-Hydroxysemicarbazide compounds **13w**-**y** and **14d**-**f** were prepared by catalytic hydrogenation of *O*-benzylated compounds **13t**-**v** i **14a**-**c** with Pd/C. Ketoprofen benzotriazolide (**10d**) reacted with several ureidoamides (**4b**,**i**,**j**,**m**) and gave esters with two molecules of ketoprofen combined covalently in one („twin drugs“).

The compounds were evaluated for their biological activity *in vitro*: antitumor, antimicrobial, antiviral and antioxidant activity, inhibition of lipoxygenase and linoleic acid lipid peroxidation. The best antitumor activity was exerted by **6l**, antiviral by **13a**,**g**,**m**, antioxidant by compound **9**. The strongest inhibitor of lipoxygenase was **13s**. Compounds **13c**,**l**,**t** showed the highest inhibition of linoleic acid lipid peroxidation. It is worth to note that compounds with the highest biological activity contained benzhydryl substituent.

Keywords: amino acid/hydroxyurea/nonsteroidal anti-inflammatory drug/semicarbazide/ester/synthesis/biological activity

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TEMELJNA DOKUMENTACIJSKA KARTICA

BASIC DOCUMENTATION CARD

KRATICE

AAPH	2,2'-azobis(2-amidinopropan) dihidroklorid
AKR	Aldo-keto reduktaza
3-AP	triapin
ATCC	American Type Culture Collection
Bn	benzil
BtcCl	klorid 1-benzotriazolkarboksilne kiseline
BtH	benzotriazol
CDI	1,1'-karbonildiimidazol
CEM	stanična linija zloćudno preobraženih T-limfocita
CLSI	Clinical and Laboratory Standards Institute
COX	ciklooksigenaza
CRFK	Crandell Rees stanice mačjeg bubrega
DCC	*N*,*N'*-dicikloheksilkarbodiimid
DMF	*N*,*N*-dimetilformamid
DSC	*N*,*N'*-disukcinimidil karbonat
DPPH	1,1-difenil-2-pikrilhidrazil radikal
EC_{50}	efektivna koncentracija pri kojoj je stvaranje plakova virusa smanjeno za 50 %
Fen	fenoprofen
GIT	gastrointestinalni trakt
H 460	stanična linija karcinoma pluća
HCT 116	stanična linija karcinoma debelog crijeva
HEL	ljudske embrionalne stanice pluća
HeLa	stanična linija karcinoma grlića maternice
Hep G2	stanična linija karcinoma jetre
HSV-1 (KOS)	herpes simpleks virus tip 1
HSV-2 (G)	herpes simpleks virus tip 2
HU	hidroksiurea
Ibu	ibuprofen
IC_{50}	koncentracija spoja koja inhibira rast stanica za 50 %
IR	infracrveno elektromagnetsko zračenje

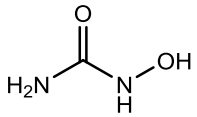
L1210	stanična linija leukemije porijeklom iz miša
LO	lipooksigenaza iz soje
5-LOX	5-lipooksigenaza
LP	lipidna peroksidacija
MCC	minimalna citotoksična koncentracija
MCF-7	stanična linija karcinoma dojke
MDCK	Madin-Darby stanice psećeg bubrega
MeKet	2-(3-benzilfenil)propanska kiselina
MiaPaCa-2	stanična linija karcinoma gušterače
MIC	minimalna inhibitorna koncentracija
MMcC	minimalna mikrobicidna koncentracija
MOLT-4	stanična linija akutne limfoblastične leukemije
Molt4/C8	stanična linija zloćudno preobraženih T-limfocita
NCCLS	National Committee for Clinical Laboratory Standards
NDGA	nordihidrogvajaretična kiselina
NMR	nuklearna magnetska rezonancija
NSAID	nesteroidni protuupalni lijek
PC-3	stanična linija karcinoma prostate
Pd/C	paladij na ugljenu
RR	ribonukleotid reduktaza
s.t.	sobna temperatura
SW620	stanična linija karcinoma debelog crijeva
TEA	trietilamin
TMS	tetrametilsilan
t_t	temperatura taljenja
WI 38	stanična linija normalnih fibroblasta porijeklom iz čovjeka

1. UVOD

1.1. OBRAZLOŽENJE TEME

1.1.1. Hidroksiuree i srodni spojevi

N-Hidroksiurea (HU, Slika 1) je citostatik koji se već dugi niz godina koristi u liječenju melanoma, kronične mijelocitne leukemije, neoperabilnog karcinoma jajnika te u lokalnoj kontroli primarnih karcinoma skvamoznih (epidermalnih) stanica glave i vrata (Lemke *et al*, 2008).



Slika 1. Hidroksiurea.

N-Hidroksiurea je također učinkovita u terapiji srpaste anemije i HIV infekcije, a koristi se samostalno ili u kombinaciji s poznatim antiretrovirusnim lijekovima. Nadalje, istražuje se njezina djelotvornost u terapiji trombocitopenije, policitemije vere, psorijaze, beta-talasemije i reumatoidnog artritisa (Mai *et al*, 2010; Navarra *et al*, 1999; Leavell *et al*, 1970).

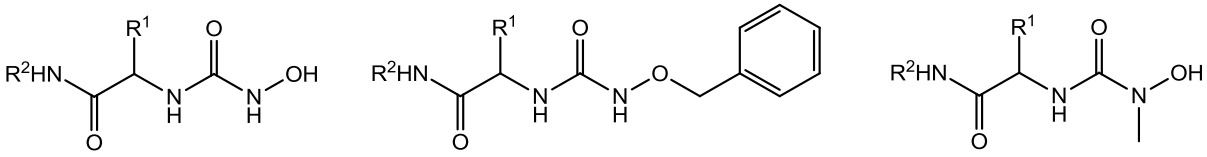
Najvažniji nedostatak hidroksiuree je kratko poluvrijeme eliminacije (1,9-3,9 h) koje je posljedica relativno male molekulske mase (M_r = 76,06) i velike polarnosti. Zbog toga se HU upotrebljava u visokim dozama (80 mg/kg svaki treći dan ili 20-30 mg/kg dnevno). Drugi važni nedostatak je brzi razvoj otpornosti na terapiju (Mai *et al*, 2010; Ren *et al*, 2002).

HU inhibira ribonukleotid reduktazu (RR, EC 1.17.4.1.), enzim koji katalizira redukciju ribonukleotida u deoksiribonukleotide te tako osigurava građevni materijal za *de novo* sintezu DNA u svim stanicama koje imaju sposobnost dijeljenja. Osim toga RR igra važnu ulogu u karcinogenezi (Szekeres *et al*, 1997; Nocentini, 1996). Zbog svojstva kompleksiranja metala HU ima inhibitorni učinak na hidrolitičke enzime koji u sebi sadrže metale: ureaze (Uesato *et al*, 2002; Fishbein *et al*, 1965), karboksipeptidazu A (Chung *et al*, 2001), ribonukleaze (Higgin *et al*, 2003) i 5-lipooksigenazu (Connolly *et al*, 1999; Kolasa *et al*, 1997; Adams *et al*, 1996; Kolasa *et al*, 1996).

HU se može smatrati derivatom hidroksamske kiseline. U hidroksamske kiseline ubrajamo neke analgetike i antiinflamatorike (ibuproksam, oksametacin, bufeksamat),

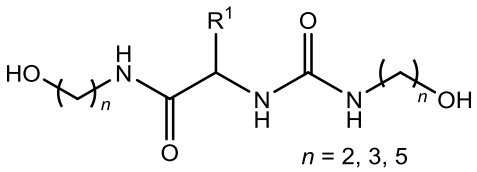
citostatike (vorinostat, didoks), antiastmatike (zileuton, atreleuton), kelatore željeza (desferioksamin), α-adrenergike i antidepresive (adrafinil).

Tijekom prijašnjih istraživanja na Zavodu za farmaceutsku kemiju sintetizirana je serija nesupstituiranih derivata hidroksiurea kojima je ispitano antitumorsko i antivirusno djelovanje (Opačić *et al*, 2005). Neki od derivata pokazali su relativno dobro antitumorsko djelovanje te su zato u okviru ovog rada pripravljeni njihovi strukturni analozi, *N*- i *O*-supstituirane hidroksiuree te im je ispitano biološko djelovanje (Slika 2) (Perković *et al*, 2008).



Slika 2. a) Nesupstituirane; b) *O*-benzilne; c) *N*-metilne hidroksiuree.

Kao nastavak ovih istraživanja, sintetizirana je serija ureidoamida (Slika 3), strukturnih analoga hidroksiurea, kod kojih je hidroksilna skupina udaljena od dušika za dva, tri ili pet ugljikovih atoma. Ti derivati su također biološki ispitani te im je djelovanje uspoređeno s hidroksiureama (Perković *et al*, 2010).



Slika 3. Ureidoamidi.

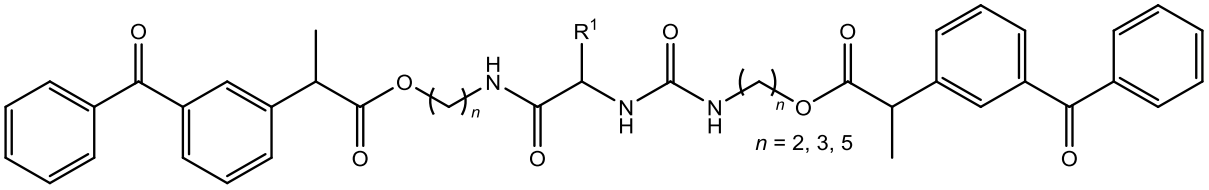
1.1.2. Derivati NSAID

Nesteroidni protuupalni lijekovi (*nonsteroidal antiinflammatory drug*, NSAID), jedni od najčešće upotrebljavanih lijekova danas, svoje djelovanje temelje na inhibiciji ciklooksigenaze (COX), enzima uključenog u sintezu endogenih eikozanoida. Aktivacijom fosfolipaze-A_2 dolazi do otpuštanja arahidonske kiseline koja se metabolizira pomoću dva enzima, ciklooksigenaze (put stvaranja prostaglandina, prostaciklina i tromboksana) i 5-lipooksigenaze (put stvaranja leukotriena) (Lemke *et al*, 2008). Inhibicija ciklooksigenaze

nesteroidnim protuupalnim lijekovima dovodi do preusmjeravanja metabolizma arahidonske kiseline preko 5-lipooksigenaze, što ima za rezultat povećanje sinteze leukotriena (pojava upale i neželjenih učinaka NSAID) i smanjene sinteze gastroprotektivnih prostaglandina (gastrointestinalne nuspojave) (Charlier *et al*, 2003; Muri *et al*, 2002).

Kemijskim modificiranjem NSAID željelo se postići nekoliko ciljeva. Smatra se da gastrointestinalnim nuspojavama doprinosi akumulacija kiselih lijekova u želucu nakon oralne primjene te inhibicija COX-1 (konstitutivna izoforma ciklooksigenaze odgovorna za sintezu gastroprotektivnih prostaglandina, protok krvi kroz bubrege i agregaciju trombocita). S druge strane, inhibicija COX-2 je poželjna jer se ona povezuje s protuupalnim djelovanjem NSAID (inducibilna izoforma ciklooksigenaze, javlja se u upaljenom tkivu, a konstitutivna je samo u bubrezima i reproduktivnim organima, povezuje se i s nekoliko tipova karcinoma, Alzheimerovom i Parkinsonovom bolesti) (Charlier *et al*, 2003). Zaštitom karboksilne skupine dobiveni su derivati s očuvanim protuupalnim učinkom, ali i novim učincima (smanjeni ulcerogeni učinak, selektivnost za COX-2, inhibicija 5-lipooksigenaze, dualno COX/5-LOX djelovanje, antitumorsko, anivirusno i antimikrobno djelovanje, smanjena lipidna peroksidacija). Općenito se smatra da bi tvari koje inhibiraju COX i 5-LOX mogle imati manje nuspojava uz očuvanu aktivnost (Liu *et al*, 2011; Fogli *et al*, 2010; Fun *et al*, 2009; Amir *et al*, 2007; Metwally *et al*, 2007; Gobec *et al*, 2005; Amir *et al*, 2004; Omar *et al*, 1996).

Najjednostavniji primjer kemijske modifikacije karboksilne skupine u svrhu smanjenja ulcerogenog učinka je esterifikacija pa je u okviru ovog rada pripravljena i serija estera NSAID (ketoprofena) s ureidoamidima gdje su na jednu molekulu ureidoamida vezane dvije molekule NSAID (Slika 4) i to pojednostavljenom metodom koja je prethodno razvijena na Zavodu za farmaceutsku kemiju (Zorc *et al*, 1994; Zorc *et al*, 1993). Može se reći da su dobiveni esteri „dvojni lijekovi“ jer sadrže dvije identične farmakoforne skupine povezane kovalentnom vezom u jednu molekulu (Huzjak *et al*, 2008). Pojam dvojnih lijekova u skupini NSAID je već poznat. Autori Fadl i Omar načinili su seriju esterskih derivata paracetamola i nekoliko NSAID sa slobodnom karboksilnom skupinom koji su bili stabilni na pH 7,4. Također, imali su analgetski učinak, bolju biodostupnost i slabiji ulcerogeni učinak od ekvivalentne smjese samih lijekova (Fadl *et al*, 1998). Biološka ispitivanja sintetiziranih esterskih derivata nadilaze okvire ovoga rada.



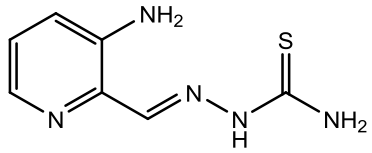
Slika 4. Esteri ketoprofena i ureidoamida.

Mnogo je dokaza koji govore u prilog tvrdnji da su NSAID potencijalni antitumorski lijekovi. Brojne epidemiološke i eksperimentalne studije potvrdile su da NSAID sprečavaju pojavu i rast tumora i da je njihova dugotrajna primjena povezana sa smanjenim rizikom od pojave karcinoma debelog crijeva, mokraćnog mjehura, jednjaka, pluća, jajnika, prostate, želuca, jetre, gušterače i jezika (Sivak-Sears *et al*, 2004; Thun *et al*, 2002). Točan mehanizam odgovoran za antitumorsko djelovanje NSAID još nije u potpunosti razjašnjen. Smatra se da su ključni inhibicija proizvodnje prostaglandina (upalni PGE_2 ima glavnu ulogu u promoviranju rasta tumora) i aktivnosti COX-2 (Wang *et al*, 2010; Wang *et al*, 2006). Poznato je da upala igra važnu ulogu u inicijaciji, progresiji i prognozi karcinoma. Nekoliko je razloga tome. Kronična upala je karakterizirana infiltracijom stanica imunološkog sustava (makrofaga, limfocita i plazma stanica), oštećenjem tkiva, fibrozom te povećanom angiogenezom, učestalim oštećenjem genoma, povećanom sintezom DNA i staničnom proliferacijom, prekidom popravaka DNA i inhibicijom apoptoze, a ti procesi su povezani i s inicijacijom i progresijom karcinoma (Rayburn *et al*, 2009). Osim toga, u kanceroznom i prekanceroznom tkivu nađena je pretjerana ekspresija COX-2. Aktivnost COX-2 utječe i na angiogenezu u tumorima te potiče njihovu progresiju aktivirajući brojne signalne puteve (Sivak-Sears *et al*, 2004).

Nadalje, NSAID inhibiraju aldo-keto reduktazu (AKR1C3) koja slabo aktivne hormone estron i androstendion reducira u aktivne 17β-estradiol, odnosno testosteron. Takvi inhibitori bi mogli naći primjenu u liječenju hormon-ovisnih oblika karcinoma prostate, dojki i endometrija (Gobec *et al*, 2005).

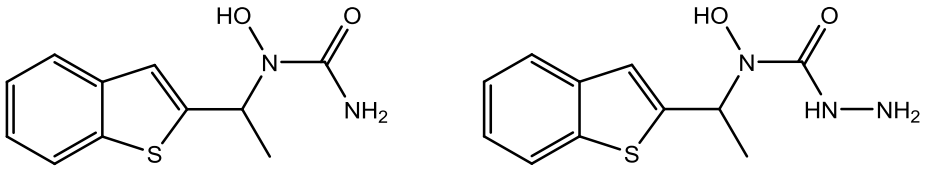
Semikarbazidi i semikarbazoni su tvari raznolike primjene. Derivati su uree koja ima važnu ulogu u metabolizmu tvari koje sadrže dušik, koriste se kao reagensi za UV detekciju α-keto kiselina u tankoslojnoj kromatografiji, a u novije vrijeme su prepoznati kao farmakofori u otkrivanju novih lijekova raznolike biološke aktivnosti. Manje su istraženi od njihovih srodnika tiosemikarbazida i tiosemikarbazona koji su pokazali antivirusno, antitumorsko i antimikrobno djelovanje (Sayin *et al*, 2010; Beraldo *et al*, 2004). Jedan

predstavnik tiosemikarbazona, triapin (3-aminopiridin-2-karboksaldehid tiosemikarb-azon, 3-AP, Slika 5) snažniji je inhibitor RR od HU. Pokazao se djelotvoran *in vitro* na stanice otporne na HU i može proći krvno-moždanu barijeru te inhibirati rast L1210 stanica (leukemija) u mozgu. U kombinaciji s drugim citostaticima ima sinergistički učinak. Nalazi se u fazama I i II kliničkih ispitivanja za liječenje nekoliko vrsta tumora, sam ili u kombinaciji s drugim citostaticima (Kolesar *et al*, 2011a; Kolesar *et al*, 2011b; Finch *et al*, 2000; Beraldo *et al*, 2004).



Slika 5. Triapin.

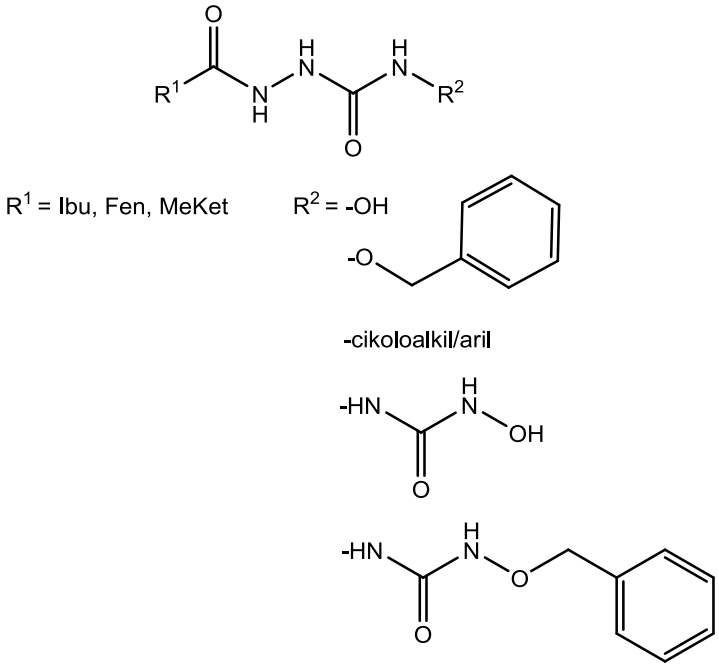
Pripravljen je semikarbazidni analog zileutona (Slika 6) te je ispitano njegovo djelovanje na inhibiciju 5-LOX. Nije se pokazao boljim inhibitorom od samog zileutona *in vitro* (Stewart *et al*, 1997).



Slika 6. a) Zileuton; b) semikarbazidni analog zileutona.

Hidroksisemikarbazidi u strukturi imaju hidroksikarbamidnu, semikarbazidnu i hidroksamsku skupinu. Zbog sličnosti s hidroksamskim kiselinama i hidroksiureama može se očekivati i slično farmakološko djelovanje (Ren *et al*, 2002).

U okviru ovog doktorskog rada pripravljena je serija semikarbazidnih/hidroksi-semikarbazidnih derivata NSAID (ibuprofena, fenoprofena i reduciranog ketoprofena) te je ispitano njihovo antitumorsko, antivirusno i antioksidativno djelovanje te njihov učinak na lipooksigenazu i lipidnu peroksidaciju linolne kiseline (Slika 7) (Perković *et al*, u tisku).



Slika 7. Semikarbazidni derivati NSAID.

1.2. LITERATURNI PREGLED

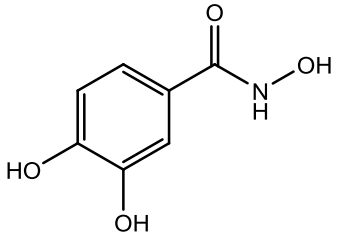
1.2.1. Hidroksiuree i srodni spojevi

1.2.1.1. *Biološko djelovanje hidroksiurea i srodnih spojeva*

Kao što je već navedeno u prethodnom poglavlju, mehanizam djelovanja HU je inhibicija ribonukleotid reduktaze čime je zaustavljeno dijeljenje stanica u ranoj S fazi staničnog ciklusa (Tomita *et al*, 1979). Osim toga, otkriveno je da HU inducira ekspresiju gena za proupalne citokine (npr. TNFα *tumor necrosis factor* i IL *interleukine*) (Navarra *et al*, 1999; Navarra *et al*, 1997).

RR se sastoji od dvije podjedinice: R1/B1 i R2/B2. R1/B1 sadrži vezna mjesta za supstrate enzima i ditiolnu skupinu koji sudjeluje u oksidoreduktivnim procesima, a R2/B2 tirozinski slobodni radikal nužan za aktivnost enzima te dva nehemska željeza koji sudjeluju u obnovi i stabilizaciji radikala (Szekeres *et al*, 1997; Nocentini, 1996). Lijekovi mogu inhibirati jednu ili obje podjedinice. HU inhibira R2/B2 podjedinicu tako da reducira tirozinski radikal (Navarra *et al*, 1999). Drugu podjedinicu inhibiraju deoksiribonukleotidi (npr. dATP, dTTP ili dGTP). Najbolji inhibitorni učinak na rast staničnih linija karcinoma (L1210-leukemija) dala je kombinacija inhibitora obje podjedinice (sinergistička inhibicija) (Szekeres *et al*, 1997).

Osim HU, još jedan derivat hidroksamskih kiselina je snažan inhibitor RR, didoks (derivat polihidroksibenzohidroksamske kiseline). On je u brojnim *in vitro* i *in vivo* istraživanjima pokazao bolju učinkovitost i manju toksičnost u usporedbi s HU (Slika 8) (Inayat *et al*, 2010; Inayat *et al*, 2002; Nocentini, 1996).



Slika 8. Didoks.

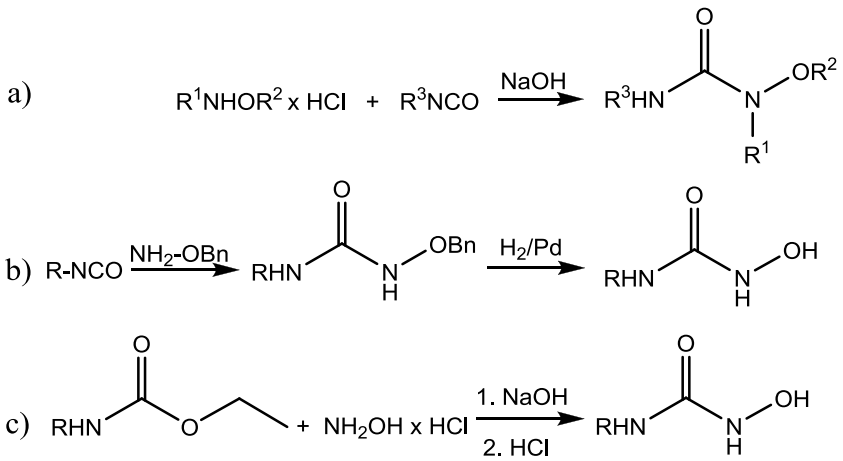
Osim inhibicije RR derivati HU pokazali su niz različitih djelovanja:

- *Inhibicija ureaze.* Ureaza katalizira hidrolizu uree do amonijaka i karbaminske kiseline koja se razlaže na još jednu molekulu amonijaka i ugljičnu kiselinu. Bakterija *Helicobacter pylori* (uzročnik gastritisa, ulkusa i karcinoma želuca) preživljava u vrlo kiselom mediju želuca upravo zbog ureaze koja razgradnjom uree iz želučanog soka povisuje pH želuca (Muri *et al*, 2004; Amtul *et al*, 2002; Odake *et al*, 1994). Tvari koje inhibiraju ureazu mogle bi biti učinkovite u liječenju infekcija s *H. pylori* (Uesato *et al*, 2002).
- *Inhibicija 5-lipooksigenaze (5-LOX).* 5-LOX je enzim koji katalizira pretvorbu arahidonske kiseline u prekursor leukotriena 5-hidroperoksieikozatetraensku kiselinu. Leukotrieni su signalne molekule uključene u procese upale i alergije (Charlier *et al*, 2003). Već je u prethodnom poglavlju naglašeno da bi tvari koje mogu inhibirati oba enzima uključena u metabolizam arahidonske kiseline (COX i 5-LOX) mogle imati bolje protuupalno djelovanje i uzrokovati manje nuspojava od konvencionalnih NSAID (Charlier *et al*, 2003; Connolly *et al*, 1999). Tako je uspoređeno djelovanje derivata atreleutona (hidroksiurea) i tepoksalina (hidroksamska kiselina). Utvrđeno je da su derivati hidroksiuree bili primarno 5-LOX inhibitori, dok su hidroksamske kiseline inhibirale oba enzima. Glavni nedostatak tepoksalina (brzi metabolizam *in vivo* do karboksilata) pokazali su i njegovi derivati (Connolly *et al*, 1999). Kod hidroksiurea derivata NSAID također se željela postići dualna inhibicija. Autori su spojili strukturu NSAID (naproksen, ibuprofen i indometacin) za koju se zna da inhibira COX s hidroksiureom koja inhibira 5-LOX te je uočen gubitak aktivnosti za COX-1 i COX-2 dok je inhibicija 5-LOX ostala očuvana (Kolasa *et al*, 1997).
- *Hipoglikemijski učinak.* Neki derivati HU snizuju razinu glukoze u krvi tako što povećavaju koncentraciju transportera za glukozu u krvi GLUT1 (Goldstein *et al*, 1993).
- Neki derivati HU pokazali su *antagonistički učinak na B_1 bradikininske receptore* za koje se zna da igraju ulogu u boli uzrokovanoj kroničnom upalom (Schnatbaum *et al*, 2010).

1.2.1.2. *Sinteza hidroksiurea*

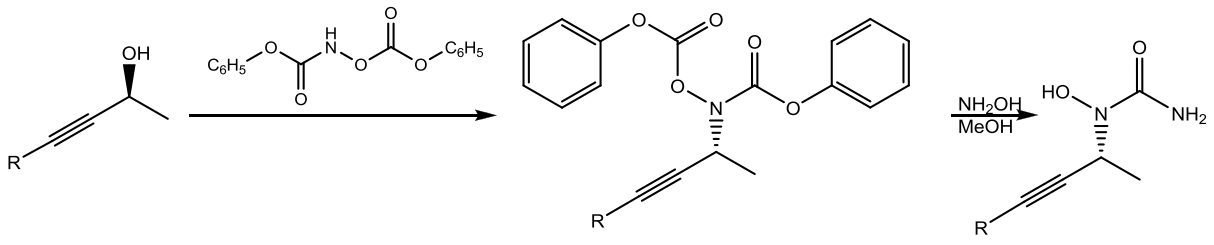
HU je prvi put sintetizirana 1869. i od tada je razvijeno mnogo metoda za sintezu njezinih derivata. Dresler i Stein su opisali sintezu tretiranjem vodene otopine hidroksilamin hidroklorida natrijevim ili drugim odgovarajućim izocijanatima (Shema 1a) (Šaban *et al*;

2009; Dresler *et al*, 1869). Tu metodu spominje i Uesato koji je u svom radu opisao sintezu niza *N*-supstituiranih HU i metodu sinteze pomoću *O*-benziloksiamina kojom se u zadnjem koraku *O*-benzilna zaštita uklanja hidrogenolizom uz 10 % Pd/C kao katalizator (Shema 1b). Za nezaštićene HU koristi metodu od Harmona i suradnika (Uesato *et al*, 2002; Harmon *et al*, 1970). Osim izocijanata, hidroksiureu daje i reakcija etil-karbamata s hidroksilamin hidrokloridom uz natrijevu lužinu (Shema 1c) (Degenghi, 1960).



Shema 1. Načini sinteze hidroksiurea

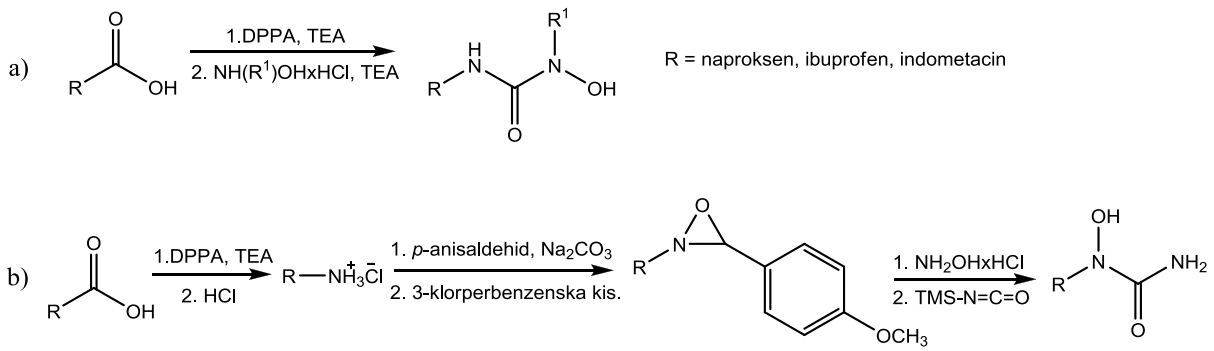
Kolasa i suradnici opisali su metodu sinteze kiralnih derivata HU iz odgovarajućeg kiralnog alkohola i *N*,*O*-bis(fenoksikarbonil)hidroksilamina, preko uretanskog međuprodukta koji je demaskiran hidroksilaminom u metanolu (Shema 2) (Kolasa *et al*, 1996; Mitsunobu, 1981).



Shema 2. Sinteza kiralnih derivata HU

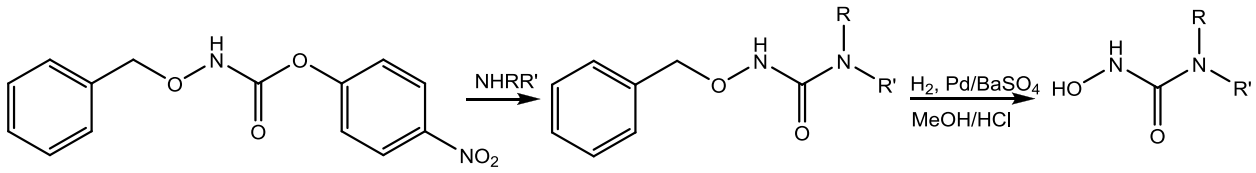
Zanimljiva je i sinteza NSAID derivata „terminalnih“ (hidroksilna skupina vezana je na dušik na koji nije vezan ostatak NSAID-a) i „unutarnjih“ (hidroksilna skupina vezana je na

dušik na koji je vezan ostatak NSAID-a) HU. Karboksilna skupina NSAID (naproksen, ibuprofen i indometacin) prevedena je pomoću difenilfosforazidata (DPPA) Curtiusovom pregradnjom u odgovarajući izocijanat koji je u reakciji s hidroksilaminom dao „terminalne“ NSAID hidroksiuree (Shema 3a). „Unutarnje“ HU dobivene su pretvorbom odgovarajućih oksaziridina u NSAID hidroksilamine i tretiranjem potonjih s trimetilsilil izocijanatom (Shema 3b) (Kürti *et al*, 2005; Kolasa *et al*, 1997).



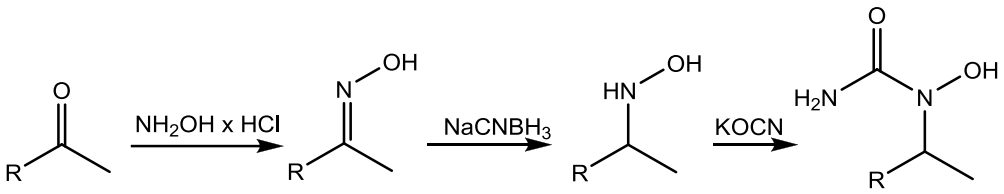
Shema 3. Sinteza NSAID hidroksiurea

Parrish i suradnici opisuju metodu sinteze derivata HU kondenzacijom različitih amina s 1-(4-nitrofenol)-*N*-(*O*-benziloksi)karbamatom uz TEA do *O*-benziliranih HU te njihovom hidrogenolizom uz $Pd/BaSO_4$ kao katalizator kako je već spomenuto (Shema 4) (Parrish *et al*, 2005).



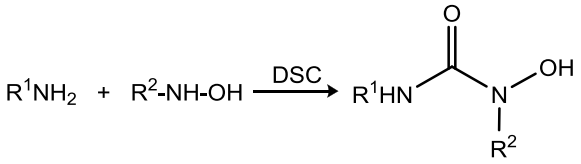
Shema 4. Sinteza derivata hidroksiurea uz hidrogenolizu

Zaitsev i suradnici započinju sintezu hidroksiurea pripravom oksima iz ketona i hidroksilamina, koji redukcijom uz cijanoborhidrid daju odgovarajuće hidroksilamine. Dalje slijedi uobičajena reakcija hidroksilamina s kalijevim izocijanatom (Shema 5) (Onuchina *et al*, 2006; Zaitsev *et al*, 2005). Na analogan način su Goldstein i suradnici dobili derivate HU i iz aldehida (Goldstein *et al*, 1993).



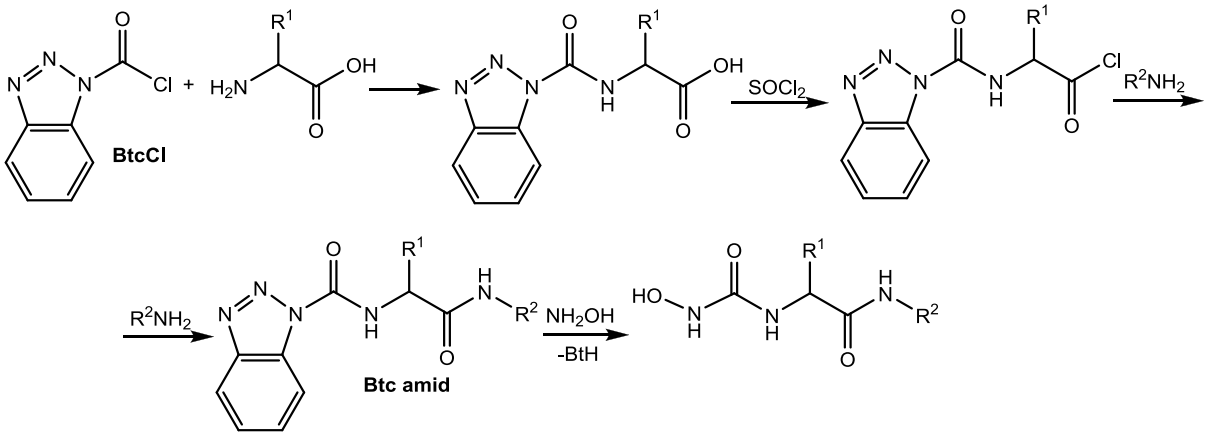
Shema 5. Sinteza hidroksiurea iz ketona

N,*N'*-disukcinimidil karbonat (DSC) je kemijski reagens koji je našao primjenu u mnogobrojnim sintezama pa tako i u sintezi hidroksiurea (Shema 6) (Schnatbaum *et al*, 2010; Ghosh *et al*, 1992).



Shema 6. Primjena DSC-a u sintezi hidroksiurea

Na Zavodu za farmaceutsku kemiju razvijena je metoda sinteze aminokiselinskih derivata hidroksiurea iz klorida 1-benzotriazolkarboksilne kiseline (BtcCl) u četiri reakcijska koraka. Zadnji korak je aminoliza amida *N*-(1-benzotriazolkarbonil)aminokiselina s hidroksilaminom (Shema 7) (Opačić *et al*, 2005).



Shema 7. Sinteza aminokiselinskih hidroksiurea

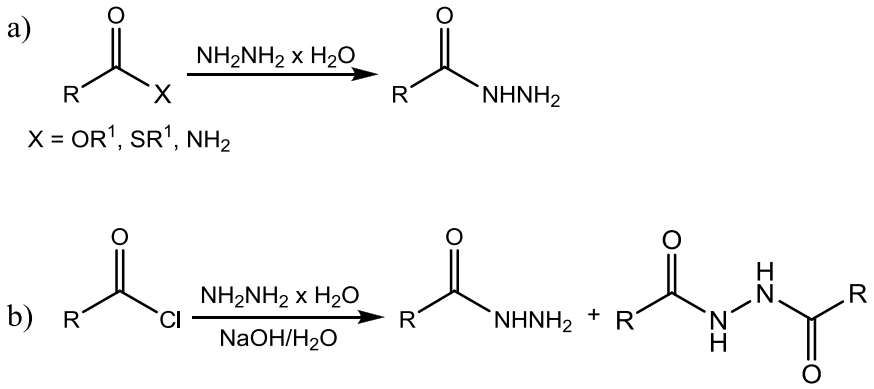
1.2.2. Biološko djelovanje i sinteza NSAID i njihovih derivata

NSAID čine skupinu lijekova različite kemijske strukture, a većina ima tri glavna tipa djelovanja: protuupalni, analgetski i antipiretski. Farmakološka aktivnost NSAID je povezana

sa supresijom biosinteze prostaglandina, prostaciklina i tromboksana iz arahidonske kiseline zbog inhibicije enzima ciklooksigenaze (Lemke *et al*, 2008). Već je spomenuto da se kao glavni uzroci gastrointestinalnih nuspojava smatraju karboksilna skupina koja je prisutna u većini NSAID-a i neselektivna inhibicija oba izoenzima koja uzrokuje manjak gastroprotektivnih prostaglandina (Charlier *et al*, 2003). Zato se većina kemijskih modifikacija na molekulama NSAID provodi upravo na karboksilnoj skupini. Osim toga, uočeno je da su derivati dobiveni zamjenom karboksilne skupine pretežno sačuvali protuupalni učinak, ali su dobili i novi (smanjeni ulcerogeni učinak, selektivnost za COX-2, inhibicija 5-LOX, dualno COX/5-LOX djelovanje, antitumorsko, anivirusno i antimikrobno djelovanje, smanjena lipidna peroksidacija) (Liu *et al*, 2011; Fogli *et al*, 2010; Fun *et al*, 2009; Amir *et al*, 2007; Metwally *et al*, 2007; Gobec *et al*, 2005; Amir *et al*, 2004; Omar *et al*, 1996). U nastavku je dan pregled sinteza i biološkog djelovanja derivata NSAID.

1.2.2.1. *Hidrazidi NSAID i derivati nastali iz hidrazida*

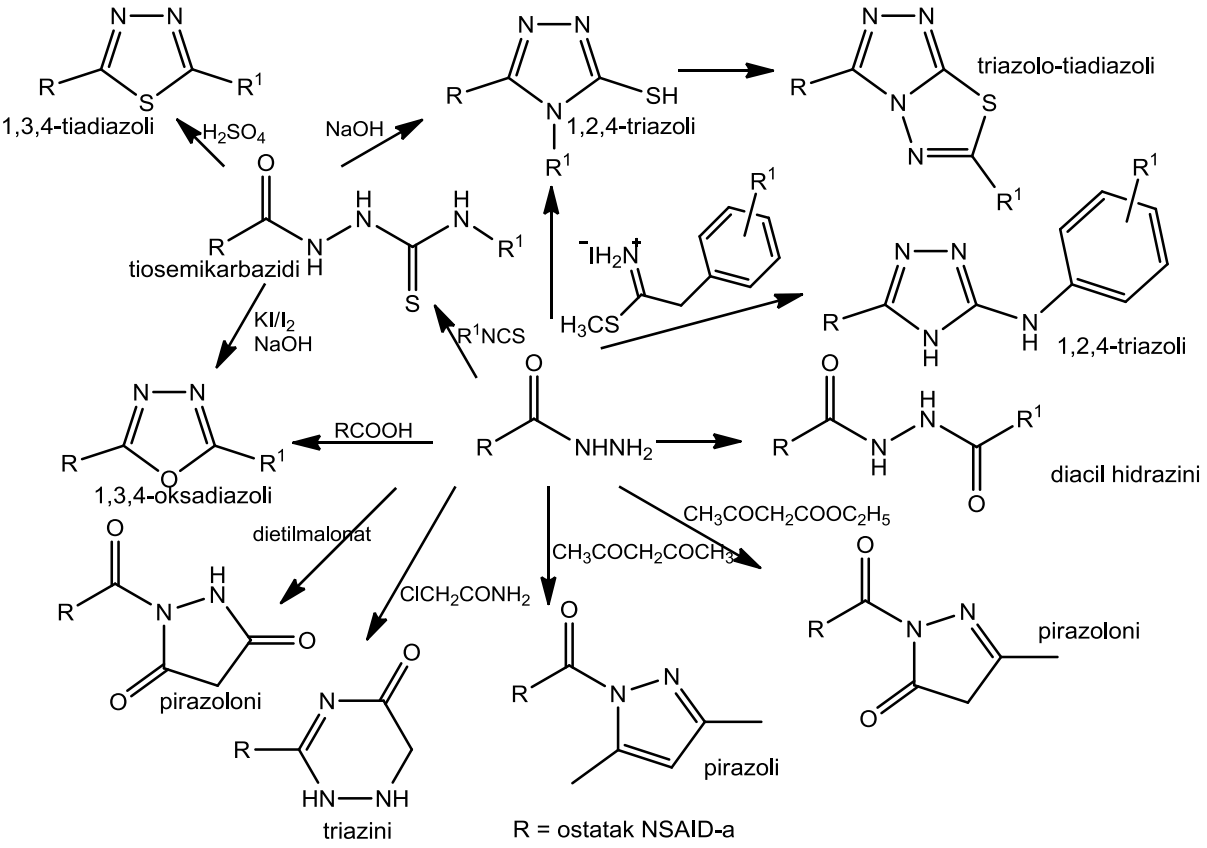
Hidrazidi ($RCONHNH_2$) se često koriste kao početni spojevi u različitim sintezama, najčešće heterocikličkih spojeva. Najčešće se dobivaju reakcijom amida, estera ili tioestera s hidrazin hidratom (Li *et al*, 2005; Edwards, 1985). Također, poznata je sinteza iz kiselinskih klorida i hidrazin hidrata, iako u toj reakciji postoji opasnost od nastajanja nusprodukta, 1,2-diacil hidrazina (Shema 8) (Li *et al*, 2005).



Shema 8. a) Nastajanje hidrazida iz estera ili amida; b) nastajanje hidrazida i diacil hidrazina iz kiselinskog klorida

Hidrazidi NSAID (ibuprofen, diklofenak, indometacin, flurbiprofen, naproksen) pronađeni u literaturi pretežno su dobiveni iz metilnih ili etilnih estera i hidrazin hidrata

refluksiranjem u etanolu 4-20 sati ili na s.t. 3 sata uz relativno dobra iskorištenja (50-80 %). Najčešće su korišteni kao početni spojevi za sintezu diacil hidrazina (Hacking *et al*, 2000) ili heterocikličkih derivata kao npr. supstituiranih pirazola (Amir *et al*, 2005), 3-supstituiranih-5-merkapto-1,2,4-triazola i 3,5-disupstituiranih-triazolo-tiadiazola (Shema 9) (Metwally *et al*, 2007; Udupi *et al*, 2000), pirazolona i 1,3,4-oksadiazola (Sharma *et al*, 2003), oksadiazola, triazina, triazola i tiadiazola (Amir *et al*, 2007; Amir *et al*, 2004; Omar *et al*, 1996).



Shema 9. Sinteza derivata NSAID iz NSAID hidrazida

Dobivenim derivatima ispitan je analgetski, protuupalni i ulcerogeni učinak te postotak lipidne peroksidacije u mukozi (Amir *et al*, 2007; Amir *et al*, 2005; Amir *et al*, 2004; Omar *et al*, 1996) (pojačana lipidna peroksidacija u želučanoj mukozi povećava udio malondialdehida koji pogoduje ulcerogenom učinku NSAID) ili akutna toksičnost (Omar *et al*, 1996). Većina ispitanih derivata imala je sačuvano protuupalno i analgetsko djelovanje, ali puno slabiji ulcerogeni učinak i lipidnu peroksidaciju u odnosu na ishodni NSAID. Neki derivati NSAID pokazali su bolji analgetski i protuupalni učinak i smanjeni ulcerogeni učinak u usporedbi s indometacinom (Metwally *et al*, 2007; Sharma *et al*, 2003).

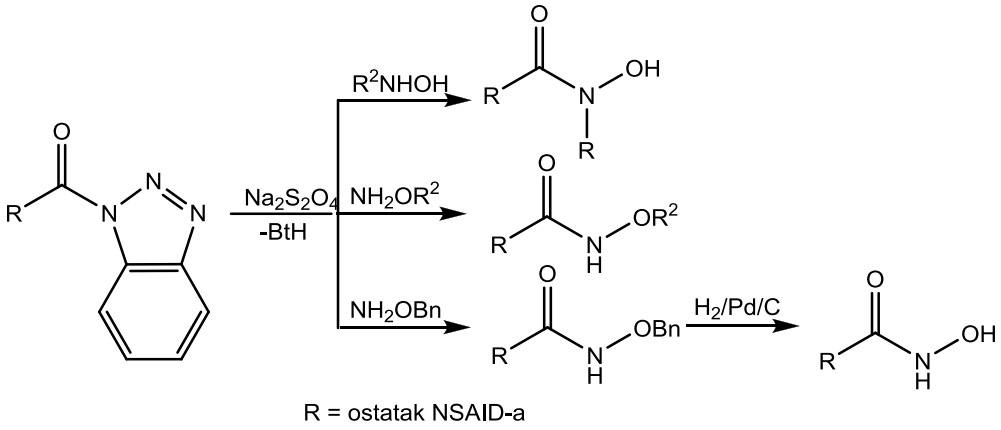
1.2.2.2. *NSAID hidroksamske kiseline i srodni spojevi*

Sinteze NSAID hidroksamskih kiselina većinom su opisane u patentima. Poznate su hidroksamske kiseline sljedećih NSAID-a: ibuprofena (ibuproksam), fenoprofena, diklofenaka, indometacina (oksametacin), meklofenaminske kiseline i sulindaka. Prvi korak u njihovoj sintezi najčešće je aktivacija karboksilne kiseline (prevođenje u ester, kiselinski klorid, anhidrid ili aktivacija pomoću CDI-a) (Kleeman *et al*, 2001; Sallmann, 1991; Cetenko, 1990; Summers *et al*, 1987; Božnar, 1987; Orzalesi, 1974; Sallmann, 1972; Marshall, 1971; Aries, 1970).

Ibuproksam i oksametacin (protuupalni lijekovi), ali i sve druge NSAID hidroksamske kiseline, imale su dualno djelovanje (inhibicija ciklooksigenaze i 5-lipooksigenaze) te su zbog toga pokazale manju akutnu toksičnost i manje nuspojava vezanih za dugotrajnu primjenu NSAID (smanjeni ulcerogeni učinak) (Muri *et al*, 2002; Flynn *et al*, 1990).

Sulindak hidroksamska kiselina i njezini sulfonski i sulfidni metaboliti pokazali su bolje *in vitro* antitumorsko (stanične linije karcinoma gušterače i debelog crijeva), proapoptotsko i antiangiogeno djelovanje u odnosu na karboksilne derivate što se objašnjava COX-ovisnim i COX-neovisnim mehanizmima. Pripravljene su iz karboksilne kiseline u reakciji s *O*-(*tert*-butil-dimetilsilil)hidroksilaminom ($TBDMSiONH_2$) u prisutnosti *N*-3-dimetilaminopropil-*N*-etilkarbodiimid hidroklorida (EDCI). Dobivenim *O*-siliranim hidroksamatima skinuta je zaštita trifluoroctenom kiselinom (Fogli *et al*, 2010).

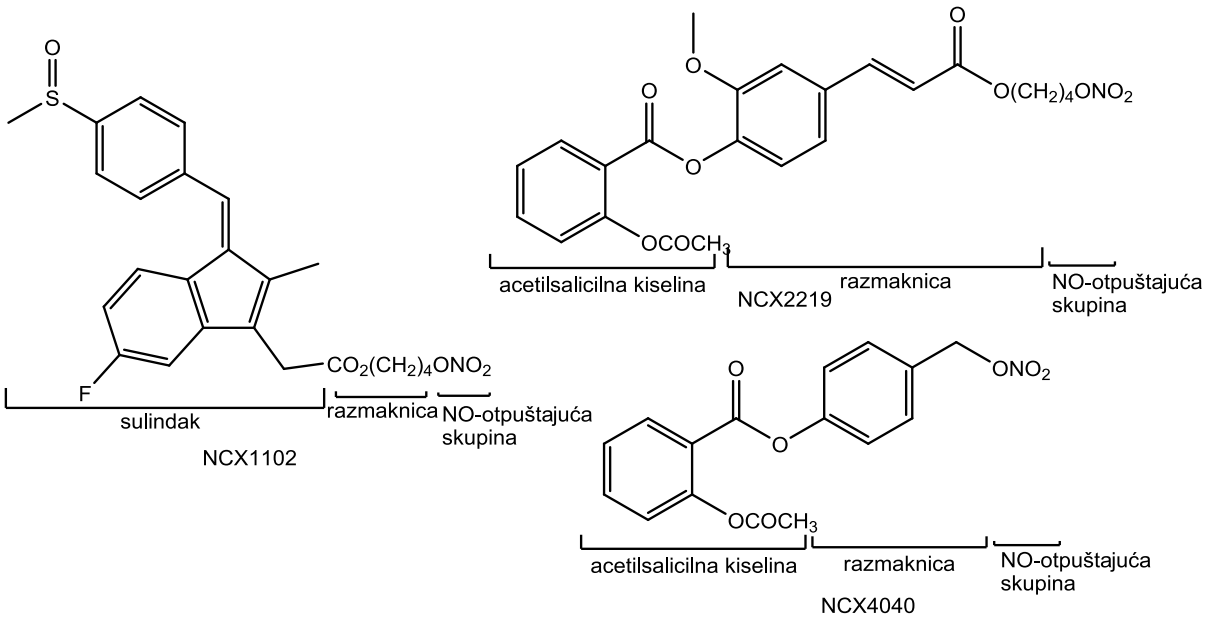
Na Zavodu za farmaceutsku kemiju sintetizirane su NSAID hidroksamske kiseline i njihovi *O*-benzil/etil/metil derivati te im je ispitano biološko djelovanje (antitumorsko, antivirusno, antimikrobno, antioksidativno, inhibicija ureaze, lipooksigenaze i lipidne peroksidacije linolne kiseline). Sintetizirani su benzotriazolskom metodom koja je korištena i u okviru ovog doktorskog rada, gdje je karboksilna skupina NSAID-a aktivirana uvođenjem benzotriazola te je nastali benzotriazolid reagirao s odgovarajućim supstituiranim ili nesupstituiranim hidroksilaminom. Nesupstituirane hidroksamske kiseline dobivene su na dva načina; u reakciji NSAID benzotriazolida s hidroksilaminom sa ili bez TEA ili katalitičkim hidrogeniranjem *O*-benziliranih hidroksamskih kiselina. Ti derivati su pokazali slabo antimikrobno i antivirusno djelovanje dok su u ostalim biološkim ispitivanjima pokazali relativno dobre rezultate (Shema 10) (Kraljević Pavelić *et al*, 2010; Rajić *et al*, 2009a; Rajić *et al*, 2009b; Zovko Končić *et al*, 2009).



Shema 10. Sinteza NSAID hidroksamskih kiselina

1.2.2.3. *NO-otpuštajući NSAID*

NSAID koji otpuštaju dušikov oksid (NO) prepoznati su kao nova klasa spojeva koja ima sačuvano protuupalno, analgetsko i antipiretsko djelovanje kao i originalni NSAID ali zbog zaštitnog djelovanja NO uzrokuju manje nuspojava u gastrointestinalnom traktu i bubrezima. Također se pokazalo da bi mogli imati i antitumorsko djelovanje što su pokazala brojna *in vitro* i *in vivo* ispitivanja (poznato je da NO inducira apoptozu u stanicama putem nekoliko različitih mehanizama). Huguenin i suradnici su tako ispitali *in vitro* antitumorski potencijal nekoliko NO-otpuštajućih NSAID-a (acetilsalicilna kiselina, indometacin, piroksikam, ibuprofen, flurbiprofen i sulindak) od kojih je najbolje djelovanje pokazao NO-sulindak (NCX 1102) te je uzet u detaljnije razmatranje (Huguenin *et al*, 2005). Andrews i suradnici usporedili su *in vitro* antitumorsko djelovanje nekoliko NSAID (*R*-flurbiprofen, sulindak-sulfon i celekoksib) i NO-acetilsalicilne kiseline na stanične linije različitih tipova karcinoma. NO-acetilsalicilna kiselina i celekoksib pokazali su značajno bolje djelovanje u odnosu na ostale ispitane spojeve. Općenito, NO-NSAID sastoje se od 3 segmenta: NSAID ostatka, razmaknice (*spacer*) i skupine koja otpušta NO (Slika 9) (Andrews *et al*, 2008).

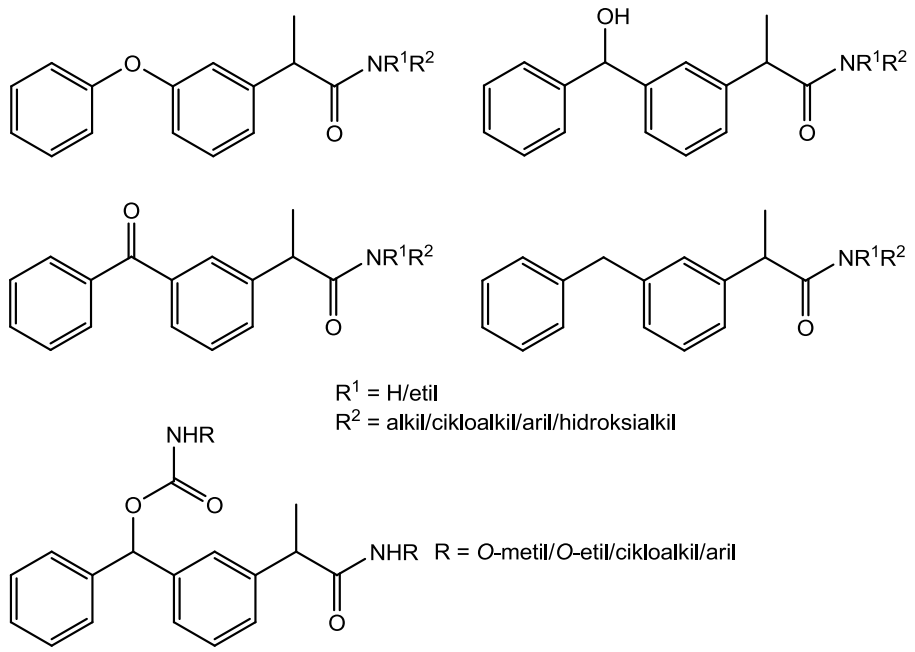


Slika 9. NO-otpuštajući NSAID.

1.2.2.4. *Amidi i esteri NSAID-a*

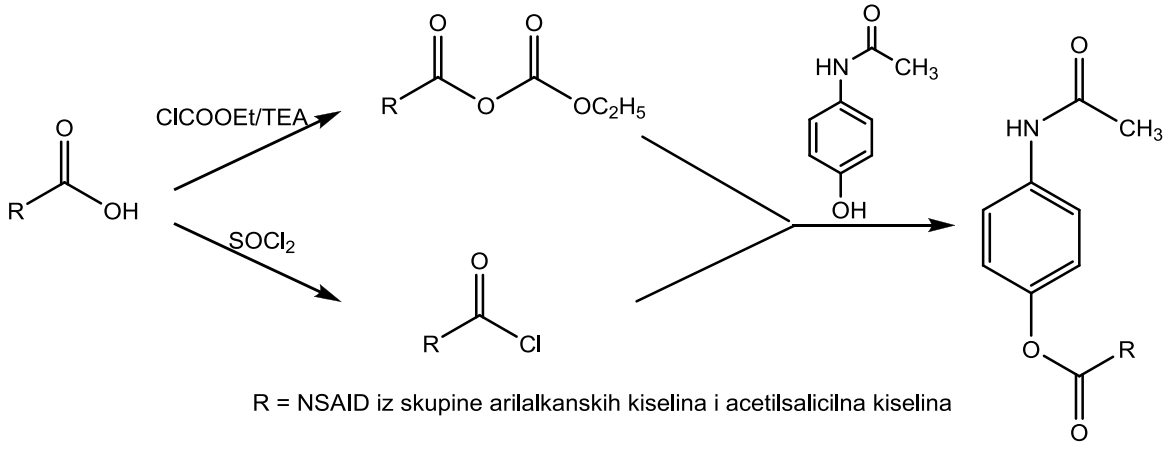
Već je puno puta spomenut štetni učinak dugotrajne primjene NSAID-a na gastrointestinalni trakt. Najjednostavniji način zaštite od učinka karboksilne skupine na mukozu je njezino privremeno maskiranje u odgovarajuće amide i estere. Takvi prolijekovi su se pokazali korisnima u smanjivanju nuspojava u odnosu na ishodne spojeve (Khan *et al*, 2006; Zhao *et al*, 2006; Shanbhag *et al*, 1992). Osim toga, modifikacijom molekule moguće je postići i neke nove učinke. Tako su npr. fenolni esteri i amidi naproksena pokazali bolje antioksidativno i antiproliferativno djelovanje od samog naproksena (Hellberg, 1999). Isto tako, koristeći nosač koji prolazi krvno-moždanu barijeru dobiveni su prolijekovi NSAID koji bi mogli biti učinkoviti u liječenju Alzheimerove bolesti (Perioli *et al*, 2004).

Neki amidi fenoprofena i ketoprofena i amidokarbamatni derivati ketoprofena (Slika 10) sintetizirani na Zavodu za farmaceutsku kemiju pokazali su jače antiproliferativno i/ili antioksidativno djelovanje ili su snažnije inhibirali LO od samih NSAID (Rajić *et al*, 2011; Rajić *et al*, 2010; Marjanović *et al*, 2007; Zovko *et al*, 2003; Zovko *et al*, 2001).



Slika 10. Amidi NSAID sintetizirani na Zavodu za farmaceutsku kemiju.

Esterifikacijom karboksilne skupine također je moguće dobiti prolijekove s poboljšanim svojstvima u odnosu na ishodni NSAID (Shanbhag, 1992). Jedan od glavnih problema u dizajniranju esterskih prolijekova je njihova slaba topljivost i brza hidroliza u GIT-u. Pripravljeni su esterski derivati ibuprofena i 1,3-dipalmitoil/distearil glicerola koji su pokazali dobru apsorpciju i hidrolizu tek pri pH 7,4 tako da je sluznica želuca ostala sačuvana (Khan *et al*, 2005). Topljivost esterskih prolijekova ibuprofena povećana je tvorbom glukopiranozidnih derivata (Zhao *et al*, 2006). Također, već su spomenuti esterski „dvojni lijekovi“ paracetamola i NSAID sa slobodnom karboksilnom skupinom i njihova poboljšana svojstva (Shema 11) (Fadl *et al*, 1998).



Shema 11. Dvojni lijekovi paracetamola i NSAID s karboksilnom skupinom

Na Zavodu za farmaceutsku kemiju pripravljeni su metilni i etilni esteri nekih NSAID (Zorc *et al*, 1993; Zorc *et al*, 1994).

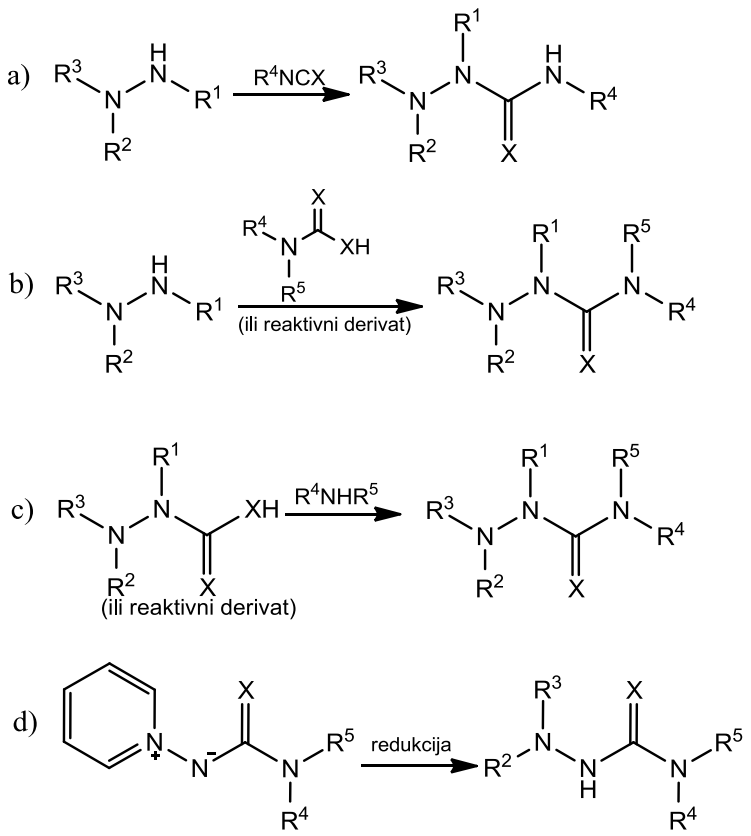
Postupci dobivanja estera i amida arilalkanskih kiselina iz skupine NSAID-a brojni su u literaturi i najčešće patentno zaštićeni. Najjednostavniji način njihovog dobivanja je prevođenje karboksilne skupine u kiselinski klorid i reakcija s alkoholom/aminom. Karboksilna skupina može se aktivirati i pomoću karbodiimida (Staab *et al*, 1998). Na Zavodu za farmaceutsku kemiju razvijen je postupak dobivanja amida i estera iz odgovarajućih benzotriazolida. Spojevi sa slobodnom karboksilnom skupinom reagiraju s kloridom 1-benzotriazolkarboksilne kiseline (BtcCl) dajući mješovite anhidride koji nakon dekarboksilacije stvaraju benzotriazolide u kojima je karboksilna skupina dovoljno aktivirana da s aminima i alkoholima daje amide i estere u vrlo blagim reakcijskim uvjetima, uz odcjepljenje benzotriazola. Amidi pretežno nastaju na sobnoj temperaturi, dok je za estere potrebna povišena temperatura (refluks) (Butula *et al*, 2007).

1.2.3. Sinteza i biološko djelovanje semikarbazida i srodnih spojeva

Već je ranije spomenuta raznolikost primjene semikarbazida i semikarbazona. Derivati su uree koja ima važnu ulogu u metabolizmu tvari koje sadrže dušik, koriste se kao reagensi za UV detekciju α-ketokiselina u tankoslojnoj kromatografiji, djeluju kao herbicidi (Hirai, 2010; Brantley, 1970), intermedijeri su u sintezi brojnih heterocikličkih spojeva (Shema 9) (Farshori *et al*, 2010) a u novije vrijeme su prepoznati kao farmakofori potencijalnih lijekova različite biološke aktivnosti (npr. antiprotozoici i antikonvulzivi) (Beraldo *et al*, 2004). Manje su istraženi od njihovih srodnika tiosemikarbazida i tiosemikarbazona koji su pokazali antivirusno, antitumorsko i antibakterijsko i antifungalno djelovanje (Sayin *et al*, 2010; Beraldo *et al*, 2004).

U literaturi se najčešće spominje postupak dobivanja semikarbazida iz odgovarajućih hidrazina i izocijanata (Shema 12a) (Brantley, 1970; Baker, 1966). U patentu US 4,725,608 opisano je još nekoliko načina dobivanja semikarbazida:

- hidrazin + karbamat (Shema 12b)
- hidrazin karboksilna kiselina (ili reaktivni derivat) + amin (Shema 12c)
- redukcija piridinium-1-il uree (Shema 12d) (Nakaguchi, 1988)



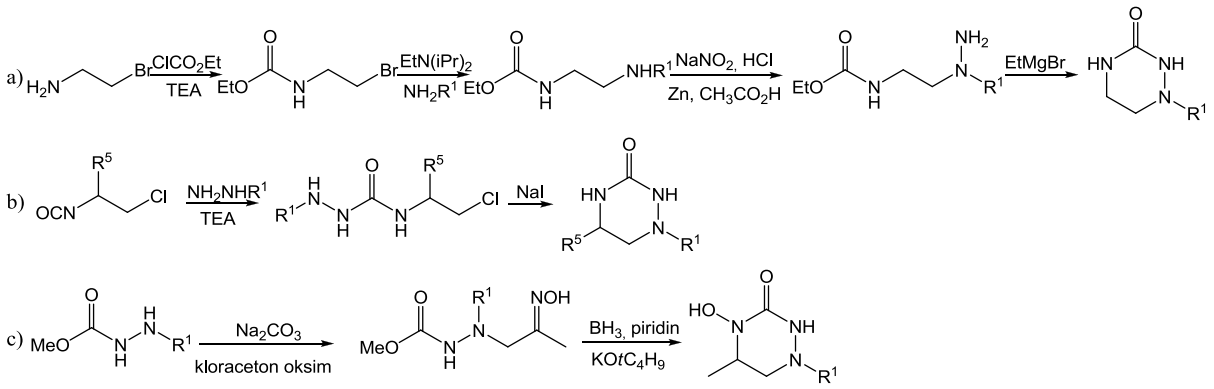
Shema 12. Sinteza semikarbazida

Neki od spomenutih semikarbazida pokazali su antiinflamatorno i analgetsko djelovanje (Nakaguchi, 1988).

Benzotiadiazinski semikarbazidi sintetizirani su iz tioetera i hidroklorida semikarbazida uz TEA (Pomarnacka *et al*, 2003). Zanimljiva je sinteza cikličkih semikarbazida 1,2,4-triazin-

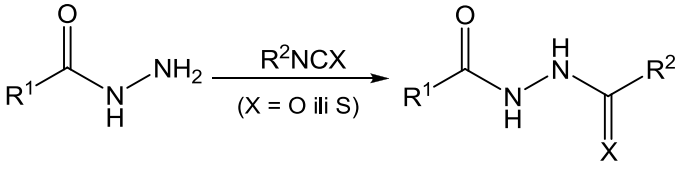
-3-ona koji su pokazali selektivno djelovanje na 5-LOX. U pronađenoj literaturi opisano je nekoliko načina njihovog dobivanja:

- iz brometilamina preko etoksikarbamata i intramolekularne ciklizacije (Shema 13a).
- iz 2-kloretil izocijanata i odgovarajućeg hidrazina; nastali produkt ciklizira na povišenoj temperaturi u DMF-u uz NaI (Shema 13b).
- *N*-hidroksi triazinoni (ciklički semikarbazidi) mogu se dobiti iz karbamata, preko odgovarajućeg oksima, koji je reduciran do hidroksilamina i cikliziran obradom s kalijevim *tert*-butoksidom (Shema 13c) (Bhatia *et al*, 1996).



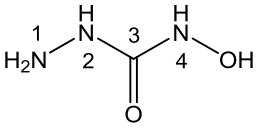
Shema 13. Sinteza cikličkih semikarbazida

Ako umjesto hidrazina, izocijanat ili izotiocijanat reagira s hidrazidima nastaju semikarbazidi ili tiosemikarbazidi s dodatnim karbonilom u strukturi, koji zatim u zadanim uvjetima ciklziraju u peteročlani prsten oksadiazol ili tiadiazol (Shema 16) (Asghar *et al*, 2010; Farshori *et al*, 2010; El-Sabbagh *et al*, 2009). Aciklički derivati (semikarbazidi i tiosemikarbazidi) pokazali su jače citotoksično djelovanje na ispitane stanične linije nekih tumora od njihovih cikličkih analoga (El-Sabbagh *et al*, 2009). Široki spektar mogućnosti ciklizacija iz semikarbazidnih i tiosemikarbazidnih derivata NSAID-a dan je u prethodnom poglavlju (poglavlje 1.2.2.1., Shema 9).



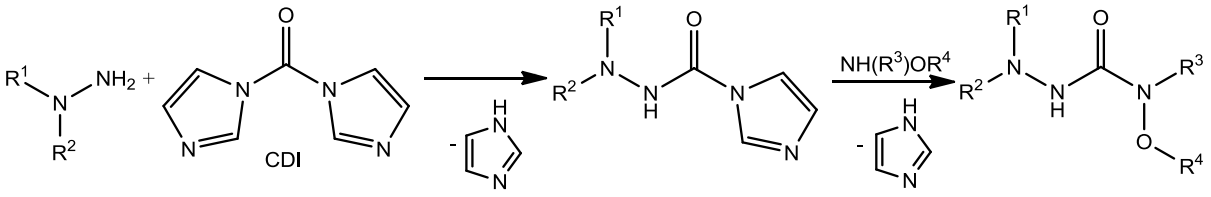
Shema 16. Sinteza karbonilsemikarbazida

4-Hidroksisemikarbazidi (Slika 11) također se koriste kao intermedijeri u sintezi cikličkih spojeva (*O*-supstituiranih triazolinona) (Müller, 1996), ali i kao spojevi s potencijalnim biološkim djelovanjem. Najčešće se spominju kao potencijalni antitumorski lijekovi zbog hidroksamske skupine koja je prisutna u cijelom nizu spojeva s antitumorskom aktivnošću (Ren *et al*, 2002).



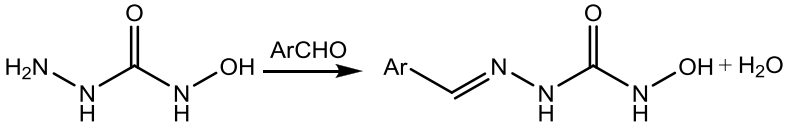
Slika 11. 4-Hidroksisemikarbazid.

Neki od prvih radova u kojima se spominju 4-hidroksisemikarbazidi su iz šezdesetih godina prošlog stoljeća. Opisana je sinteza 4-hidroksisemikarbazida iz etoksikarbonil hidrazina i hidroksilamina (Zinner, 1968), *O*-metilhidroksisemikarbazid dobiven je iz estera *N*-metoksikarbaminske kiseline i hidrazina (Petersen, 1972; Ohme *et al*, 1971), dok esteri karbazinske kiseline s hidroksilaminima daju 4-hidroksisemikarbazide (Shema 13c) (Bhatia *et al*, 1996). Zilz opisuje postupak sinteze 4-hidroksisemikarbazida iz supstituiranih hidrazina i hidroksilamina uz CDI (Shema 17) (Zilz, 2000).



Shema 17. Sinteza 4-hidroksisemikarbazida uz CDI

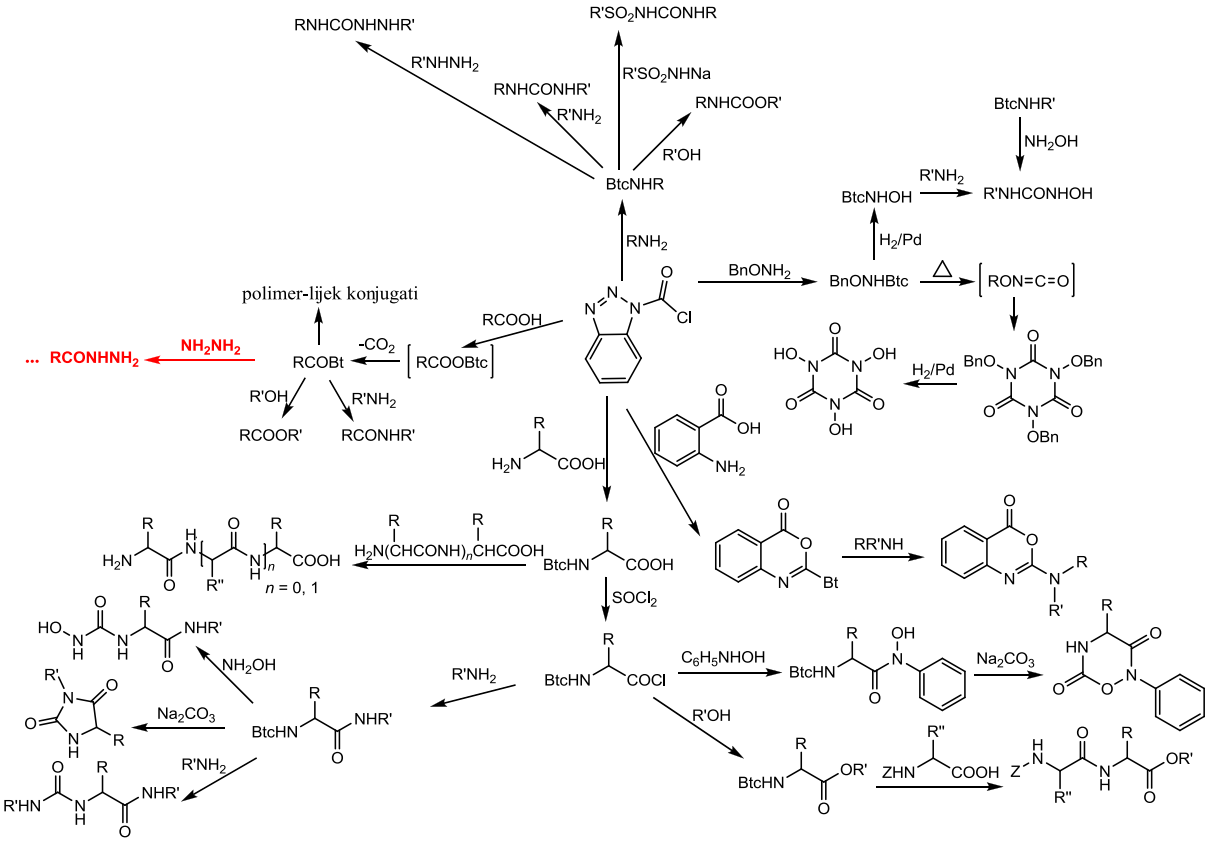
Godine 2002. skupina autora objavila je sintezu serije Schiffovih baza hidroksisemikarbazida (SB-HSC) prema modificiranom ranije opisanom postupku (Grobner *et al*, 1974; Steinberg *et al*, 1956) koji su sintetizirani iz samog 4-hidroksisemikarbazida (dobiven iz *N*-hidroksifenil karbamata i hidrazin hidrata u etanolu) i odgovarajućeg aldehida u ekvimolarnim omjerima (Shema 18) (Ren *et al*, 2002). Provedena QSAR analiza tih derivata potvrdila je da su hidroksiurea i lipofilnost važni čimbenici za antitumorsko djelovanje (Raichurkar *et al*, 2003).



Shema 18. Schiffove baze 4-hidroksisemikarbazida

1.2.4. Benzotriazol u sintetskoj kemiji

Benzotriazol (BtH) pripada u skupinu azola (aromatski peteročlani heterociklički ili benzenom kondenzirani heterociklički spojevi s dva ili više dušikova atoma, npr. imidazol, pirazol, triazol, tetrazol, benzimidazol i drugi) (Staab *et al*, 1998). Benzotriazol se u kemijskim sintezama počeo koristiti osamdesetih godina prošlog stoljeća i od tada je upotrebljen u sintezi cijelog niza spojeva. Benzotriazol se pokazao kao vrlo korisno pomoćno sredstvo u sintezama: lako se izdvaja iz reakcijske smjese te se može ponovo upotrijebiti, lako se ugrađuje u početni spoj te ga aktivira za njegovu daljnju kemijsku pretvorbu, stabilan je pri različitim kemijskim operacijama. Osim toga nije skup, dobro je topljiv u velikom broju organskih otapala i zaluženoj vodi te djelomično topljiv u vodi (Katritzky *et al*, 2010; Katritzky *et al*, 1998). Također, benzotriazol reagira s fozgenom u molarnom omjeru 1:1 dajući klorid 1-benzotriazolkarboksilne kiseline (BtcCl) koji poput običnih kiselinskih klorida daje derivate koji pokazuju osobito reaktivna svojstva. Zbog otežanog rukovanja fozgenom i sigurnosti u radu u laboratoriju, danas se u sintezi BtcCl-a pretežno upotrebljava čvrsti trifozgen (trimer fozgena) koji se zagrijavanjem razlaže u fozgen (Shema 20) (Kalčić *et al*, 2003; Butula *et al*, 1977). BtcCl vrlo lako reagira s nukleofilima dajući cijeli niz reaktivnih spojeva iz kojih je do sada pripravljen velik broj organskih spojeva. Neki od njih su karbamati, simetrične i asimetrične uree, semikarbazidi, karbazidi, sulfoniluree, sulfonilkarbazidi, esteri nitroalkanskih kiselina, brojni heterociklički spojevi (derivati benzoksazina, kinazolina, triazintriona, hidantoina i oksadiazina), derivati aminokiselina (esteri i amidi aminokiselina, di- i tripeptidi, hidantoinske kiseline, hidroksiuree) i derivati ljekovitih tvari. Funkcionalne skupine NSAID-a ketoprofena, fenoprofena, ibuprofena, diklofenaka i indometacina, urikozurika probenecida, antihiperlipemika gemfibrozila i estrogena estradiol-benzoata aktivirane su uvođenjem benzotriazola pomoću BtcCl-a te su dobiveni benzotriazolidi koji su s odgovarajućim reagensima dali amide, hidroksamske kiseline, estere i polimer-lijek konjugate (polimerni prolijekovi u kojima je ljekovita tvar povezana kovalentnom vezom na polimerni nosač poliaspartamidnog tipa). Pregled „benzotriazolskih“ sinteza do sada napravljenih na Zavodu za farmaceutsku kemiju dan je u Shemi 19 (Butula *et al*, 2007). Također, ovaj doktorski rad dat će uvid u daljnje mogućnosti primjene benzotriazolske kemije u sintezi organskih spojeva.



Shema 19. Raznolikost primjene benzotriazola u sintetskoj kemiji

2. MATERIJALI I METODE

2.1. SINTEZE

2.1.1. Materijali i instrumenti

Tališta (t_t) su određena na Stuart SMP3 instrumentu za određivanje tališta (Barloworld Scientific, UK) i nisu korigirana. Iskorištenja nisu optimirana. CHN analiza je provedena na CHN-LECO-932 instrumentu za elementarnu analizu (LECO Corporation, USA) te su sve elementarne analize bile unutar 0,4 %.

Za tankoslojnu kromatografiju upotrebljene su silikagel ploče 60 F_{254} (Kemika, Hrvatska), te diklormetan/metanol (9:1, 4:1, 9,5:0,5), cikloheksan/etil-acetat (1:1), petroleter/etil-acetat/metanol (3:1:0,5) i etil-acetat/petroleter/metanol (2:1:0,1) kao pokretna faza. Za kromatografiju na koloni kao nepokretna faza korišten je silikagel veličine čestica 0,063–0,200 nm (Merck, Njemačka i Kemika, Hrvatska), a cikloheksan/etil-acetat/metanol (3:1:0,6), diklormetan/metanol (9:1 i 8,5:1,5, 4:1, 9,4:0,6), kloroform/metanol (9,5:0,5) kao pokretne faze. Analizirani spojevi detektirani su UV zračenjem (λ = 254 nm), parama joda, etanolnom otopinom fosfomolibdenske kiseline i otopinom željezovog(III) klorida (w = 1 %).

IR spektri snimljeni su na Paragon FT-IR spektrofotometru (Perkin Elmer, UK). NMR spektri snimljeni su Bruker AV-600 spektrometrom (Bruker, USA) kod 600,13 ili 300,13 MHz za ^{1}H, odnosno kod 150,917 ili 75,47 MHz za ^{13}C jezgru. Uzorci su mjereni u DMSO-d_6 u NMR cjevčicama od 5 mm. Kemijski pomaci dani su u ppm u odnosu na tetrametilsilan (TMS) kao unutarnji standard. FID rezolucija u ^{1}H NMR i ^{13}C NMR spektrima bila je 0,29 Hz i 0,54 Hz po točki.

Benzotriazol, trifozgen, *N*-metilhidroksilamin hidroklorid, *O*-benzilhidroksil-amin hidroklorid, ciklopentilamin, cikloheksilamin, cikloheksanmetilamin, benzilamin, feniletilamin, benzhidrilamin, 2-aminoetanol, 3-aminopropanol, 5-aminopentanol L-alanin i 10 %-tni Pd/C nabavljeni su od tvrtke Aldrich (Njemačka), TEA od Sigme (USA), L-valin, L-leucin i L-fenilalanin od Kemike (Hrvatska). D-fenilglicin i NSAID-i (ketoprofen, fenoprofen i ibuprofen) dobiveni su susretljivošću tvornica lijekova Pliva i Belupo (Hrvatska) i Sveučilišta u Potchefstroomu (Južnoafrička Republika).

U eksperimentalnom dijelu korištena su bezvodna otapala. Bezvodni toluen dobiven je sljedećim postupkom: toluen je ekstrahiran vodom, zatim je osušen nad bezvodnim kalcijevim kloridom, destiliran i čuvan nad elementarnim natrijem. Bezvodni eter priređen je na sljedeći način: eter je ekstrahiran 3 puta otopinom kalcijevog klorida (w = 10 %), 1 put otopinom sumporne kiseline (w = 5 %), 1 put vodom, 1 put otopinom natrijevog karbonata

(w = 5 %) i 1 put otopinom željezovog(II) sulfata (w = 5 %). Nakon toga eter je opran do neutralnog pH, sušen nad bezvodnim kalcijevim kloridom 24 h, predestiliran i čuvan nad elementarnim natrijem. Bezvodni dioksan: dioksan je refluksiran 24 h nad elementarnim natrijem, predestiliran i čuvan nad elementarnim natrijem.

Sve ostale kemikalije bile su *p. a*. čistoće.

2.1.2. Sinteza klorida 1-benzotriazolkarboksilne kiseline (1)

Otopina BtH (1,191 g, 10 mmol) i trifozgena (2,523 g, 8,5 mmol) u bezvodnom toluenu (20 mL) refluksirana je 3 h. Reakcijska smjesa uparena je pod sniženim tlakom i nekoliko puta naparena toluenom. Dobiveni klorid 1-benzotriazolkarboksilne kiseline (BtcCl, **1**) upotrebljen je u daljnjim reakcijama bez čišćenja.

2.1.3. Sinteza *N*-(1-benzotriazolkarbonil)aminokiselina 2a-e. Opća metoda

Suspenziji odgovarajuće aminokiseline (20 mmol) u bezvodnom dioksanu (40 mL) uz miješanje je dokapana otopina BtcCl (**1**) (1,810 g, 10 mmol) u bezvodnom dioksanu (10 mL) tijekom 0,25 h. Nakon dokapavanja reakcijska smjesa miješana je 24 h na sobnoj temperaturi. Talog hidroklorida aminokiseline je odsisan, a otapalo upareno pod sniženim tlakom. Dobivena sirova *N*-(1-benzotriazolkarbonil)aminokiselina je zatim isprana vrućim toluenom (**2a**, **2b**, **2e**) ili prekristalizirana iz toluena (**2c** i **2d**). IR spektri i tališta spojeva **2a-e** u potpunosti odgovaraju ranije sintetiziranim spojevima (Butula *et al*, 1981a).

2.1.4. Sinteza klorida *N*-(1-benzotriazolkarbonil)aminokiselina 3a-e. Opća metoda

Otopina odgovarajuće *N*-(1-benzotriazolkarbonil)aminokiseline **2a-e** (4 mmol) u tionil-kloridu (20 mL) miješana je na sobnoj temperaturi 24 h. Tionil-klorid uparen je pod sniženim tlakom, a dobiveni ostatak nekoliko puta naparen bezvodnim toluenom. Dobiveni kloridi **3a-e** upotrebljeni su u daljnjim reakcijama bez čišćenja.

2.1.5. Sinteza ureidoamida 4a-o. Opća metoda

U otopinu klorida *N*-(1-benzotriazolilkarbonil)-aminokiseline (**3a-e**) (1 mmol) u dioksanu dodana je otopina 0,183 g (3mmol) 2-aminoetanola ili 0,225 g (3 mmol) 3-

aminopropanola ili 0,309 g (3 mmol) 5-aminopentanola u dioksanu. Reakcijska smjesa je miješana na sobnoj temperaturi 24 sata te je uparena pod sniženim tlakom. Čisti produkt dobiven je kromatografijom na koloni te prekristalizacijom.

1-(1-(2-Hidroksietilkarbamoil)etil-3-(2-hidroksietil)urea (4a)

Količina reaktanta: 0,253 g spoja **3a**. Sirovi produkt **4a** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklormetan/metanol 8,5:1,5 → 4:1 i rastrljavanjem u eteru.

Iskorištenje: 0,132 g (61 %).

t_t 150–154 °C.

IR (KBr): ν_{max} 3422, 3274, 2969, 2937, 2879, 1644, 1570, 1056 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,87 (t, 1H, 1'(amid), J = 5,4 Hz), 6,21 (d, 1H, 3, J = 7,9 Hz), 6,14 (t, 1H, 1'(urea), J = 5,5 Hz), 4,69, 4,67 (2t, 2H, 4', J = 5,6 Hz, J = 5,2 Hz), 4,16–4,06 (m, 1H, 2), 3,38–3,32 (m, 4H, 3'), 3,13–3,00 (m, 4H, 2'), 1,12 (d, 3H, 5, J = 7,4 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,78 (1), 157,98 (4), 61,19, 60,21 (3'), 49,07 (2), 42,46, 41,86 (2'), 20,11 (5).

1-(1-(3-Hidroksipropilkarbamoil)etil-3-(3-hidroksipropil)urea (4b)

Količina reaktanta: 0,253 g spoja **3a**. Sirovi produkt **4b** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklormetan/metanol 4:1 i rastrljavanjem u eteru.

Iskorištenje: 0,139 g (56 %).

t_t 152–156 °C.

IR (KBr): ν_{max} 3275, 2941, 2875, 1638, 1560, 1060 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,85 (t, 1H, 1'(amid), J = 5,5 Hz), 6,05–6,02 (d, t, 2H, 1'(urea), 3), 4,43, 4,42 (2t, 2H, 5', J = 4,8 Hz, J = 5,1 Hz), 4,14–4,05 (m, 1H, 2), 3,39 (q, 4H, 4', J = 6,1 Hz), 3,12–3,00 (m, 4H, 2'), 1,58–1,44 (m, 4H, 3'), 1,12 (d, 3H, 5, J = 6,8 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,63 (1), 157,97 (4), 58,83, 58,82 (4'), 49,02 (2), 36,70, 36,12 (2'), 33,66, 32,79 (3'), 20,23 (5).

1-(1-(5-Hidroksipentilkarbamoil)etil-3-(5-hidroksipentil)urea (4c)

Količina reaktanta: 0,253 g spoja **3a**. Sirovi produkt **4c** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklormetan/metanol 4:1 i rastrljavanjem u eteru.

Iskorištenje: 0,180 g (59 %).

t_t 142–146 °C.

IR (KBr): ν_{max} 3360, 3282, 3111, 2936, 2862, 1627, 1561, 1056 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,84 (t, 1H, 1'(amid), J = 5,3 Hz), 6,03 (t, 1H, 1'(urea), J = 5,5 Hz), 5,96 (d, 1H, 3, J = 7,8 Hz), 4,34 (t, 2H, 7', J = 5,1 Hz), 4,15–4,05 (m, 1H, 2), 3,37 (q, 4H, 6', J = 5,8 Hz), 3,05–2,92 (m, 4H, 2'), 1,45–1,29 (m, 12H, 3'–5'), 1,12 (d, 3H, 5).

^{13}C NMR (DMSO-d_6, δ ppm): 173,44 (1), 157,77 (4), 61,13, 61,08 (6'), 48,95 (2), 39,63, 38,89 (2'), 32,72, 32,63 (5'), 30,35, 29,39 (3'), 23,39, 23,30 (4'), 20,36 (5).

1-(1-(2-Hidroksietilkarbamoil)-2-metilpropil-3-(2-hidroksietil)urea (4d)

Količina reaktanta: 0,281 g spoja **3b**. Sirovi produkt **4d** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 → 4:1 i prekristalizacijom iz metanola i etera.

Iskorištenje: 0,140 g (57 %).

t_t 194–198 ºC.

IR (KBr): ν_{max} 3280, 3102, 2969, 2874, 1626, 1571, 1063 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,87 (t, 1H, 1'(amid), J = 5,4 Hz), 6,16 (t, 1H, 1'(urea), J = 5,5 Hz), 6,11 (d, 1H, 3, J = 9,1 Hz), 4,65, 4,64 (2t, 2H, 4', J = 4,8 Hz, J = 5,1 Hz), 3,97 (dd, 1H, 2, J = 6,0 Hz, J = 2,9 Hz), 3,45–3,33 (m, 4H, 3'), 3,18–3,03 (m, 4H, 2'), 1,91–1,80 (m, 1H, 5), 0,82 (d, 3H, 6, J = 6,8 Hz), 0,78 (d, 3H, 7, J = 6,8 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,60 (1), 158,44 (4), 61,25, 60,28 (3'), 58,34 (2), 42,49, 41,76 (2'), 31,59 (5), 19,73, 18,25 (6, 7).

1-(1-(3-Hidroksipropilkarbamoil)-2-metilpropil-3-(3-hidroksipropil)urea (4e)

Količina reaktanta: 0,281 g spoja **3b**. Sirovi produkt **4e** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 9:1 → 4:1 i prekristalizacijom iz metanola i etera.

Iskorištenje: 0,154 g (56 %).

t_t 178–181 ºC.

IR (KBr): ν_{max} 3350, 3279, 3102, 2963, 2874, 1630, 1566, 1234, 1078, 1044 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,90 (t, 1H, 1'(amid), J = 5,6 Hz), 6,06 (t, 1H, 1'(urea), J = 5,7 Hz), 5,96 (d, 1H, 3, J = 9,1 Hz), 4,42, 4,41 (2t, 2H, 5', J = 5,4 Hz), 3,95 (dd, 1H, 2, J = 6,1 Hz, J = 3,0 Hz), 3,39 (q, 4H, 4', J = 6,0 Hz), 3,19–3,00 (m, 4H, 2'), 1,89–1,78 (m, 1H, 5), 1,58–1,45 (m, 4H, 3'), 0,82 (d, 3H, 6, J = 6,8 Hz), 0,79 (d, 3H, 7, J = 7,0 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,40 (1), 158,45 (4), 58,89, 58,79 (4'), 58,28 (2), 36,68, 36,07 (2'), 33,71, 32,86 (3'), 31,72 (5), 19,71, 18,35 (6, 7).

1-(1-(5-Hidroksipentilkarbamoil)-2-metilpropil-3-(5-hidroksipentil)urea (4f)

Količina reaktanta: 0,281 g spoja **3b**. Sirovi produkt **4f** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 9:1 → 4:1 i rastrljavanjem u eteru.

Iskorištenje: 0,171 g (52 %).

t_t 159–162 ºC.

IR (KBr): ν_{max} 3349, 3278, 3107, 2935, 2861, 1628, 1572, 1236, 1059 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,89 (t, 1H, 1'(amid), J = 5,5 Hz), 6,05 (t, 1H, 1'(urea), J = 5,4 Hz), 5,90 (d, 1H, 3, J = 9,0 Hz), 4,35–4,32 (m, 2H, 7'), 3,95 (dd, 1H, 2, J = 6,2 Hz, J = 2,7 Hz), 3,38–3,35 (m, 4H, 6'), 3,12–2,93 (m, 4H, 2'), 1,88–1,77 (m, 1H, 5), 1,44–1,25 (m, 12H, 3'–5'), 0,81 (d, 3H, 6, J = 6,8 Hz), 0,78 (d, 3H, 7, J = 6,9 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,23 (1), 158,25 (4), 61,13, 61,08 (6'), 58,21 (2), 39,67, 38,83 (2'), 32,72, 32,62 (5'), 31,78 (5), 30,35, 29,41 (3'), 23,39, 23,35 (4'), 19,70, 18,33 (6, 7).

1-(1-(2-Hidroksietilkarbamoil)-3-metilbutil-3-(2-hidroksietil)urea (4g)

Količina reaktanta: 0,295 g spoja **3c**. Sirovi produkt **4g** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklormetan/metanol 8,5:1,5 i prekristalizacijom iz metanola i etera.

Iskorištenje: 0,138 g (53 %).

t_t 157–161 ºC.

IR (KBr): ν_{max} 3290, 3098, 2954, 2928, 2872, 1625, 1569, 1059 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,87 (t, 1H, 1'(amid), J = 5,3 Hz), 6,11 (d, 1H, 3, J = 8,9 Hz), 6,04 (t, 1H, 1'(urea), J = 5,4 Hz), 4,65–4,64 (m, 2H, 4'), 4,15–4,07 (m, 1H, 2), 3,40–3,34 (m, 4H, 3'), 3,13–3,01 (m, 4H, 2'), 1,60–1,52 (m, 1H, 6), 1,42–1,27 (m, 2H, 5), 0,86 (2d, 6H, 7, 8, J = 6,2 Hz, J = 5,4 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,69 (1), 158,18 (4), 61,21, 60,24 (3'), 51,95 (2), 42,84, 42,49 (2'), 41,83 (5), 24,69 (6), 23,51, 22,39 (7, 8).

1-(1-(3-Hidroksipropilkarbamoil)-3-metilbutil-3-(3-hidroksipropil)urea (4h)

Količina reaktanta: 0,295 g spoja **3c**. Sirovi produkt **4h** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklormetan/metanol 8,5:1,5 i rastrljavanjem u eteru.

Iskorištenje: 0,150 g (52 %).

t_t 109–111 ºC.

IR (KBr): ν_{max} 3308, 3098, 2957, 2872, 1630, 1572, 1252, 1073 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,89 (t, 1H, 1'(amid), J = 5,5 Hz), 5,97–5,94 (m, 2H, 3, 1'(urea)), 4,42, 4,40 (2t, 2H, 5', J = 5,3 Hz), 4,13–4,05 (m, 1H, 2), 3,42–3,35 (m, 4H, 4'), 3,15–2,99 (m, 4H, 2'), 1,59–1,44 (m, 5H, 6, 3'), 1,39–1,30 (m, 2H, 5), 0,87 (d, 3H, 7, J = 6,2 Hz), 0,85 (d, 3H, 8, J = 6,2 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,50 (1), 158,19 (4), 58,90, 58,85 (4'), 51,95 (2), 42,92 (5), 36,73, 36,11 (2'), 33,68, 32,81 (3'), 24,74 (6), 23,43, 22,49 (7, 8).

1-(1-(5-Hidroksipentilkarbamoil)-3-metilbutil-3-(5-hidroksipentil)urea (4i)

Količina reaktanta: 0,295 g spoja **3c**. Sirovi produkt **4i** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 9:1 → 8,5:1,5 i rastrljavanjem u eteru.

Iskorištenje: 0,270 g (78 %).

t_t 83–86 ºC.

IR (KBr): ν_{max} 3349, 3280, 3100, 2937, 2862, 1626, 1562, 1262, 1059 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,88 (t, 1H, 1'(amid), J = 5,4 Hz), 5,94 (t, 1H, 1' (urea), J = 5,7 Hz), 5,88 (d, 1H, 3, J = 9,0 Hz), 4,33 (t, 2H, 7', J = 5,1 Hz), 4,13–4,06 (m, 1H, 2), 3,39–3,33 (m, 4H, 6'), 3,08–2,92 (m, 4H, 2'), 1,58–1,22 (m, 15H, 6, 5, 3'–5'), 1,39–1,30 (m, 2H, 5), 0,87 (d, 3H, 7, J = 6,1 Hz), 0,85 (d, 3H, 8, J = 6,1 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,29 (1), 157,98 (4), 61,13, 61,09 (6'), 51,90 (2), 43,03 (5), 39,70, 38,87 (2'), 32,73, 32,64 (5'), 30,35, 29,37 (3'), 24,74 (6), 23,41, 22,57 (7, 8), 23,38, 23,31 (4').

1-((2-Hidroksietilkarbamoil)(fenil)metil)-3-(2-hidroksietil)urea (4j)

Količina reaktanta: 0,315 g spoja **3d**. Sirovi produkt **4j** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 → 4:1 i rastrljavanjem u eteru.

Iskorištenje: 0,171 g (61 %).

t_t 206–208 ºC.

IR (KBr): ν_{max} 3291, 3092, 2934, 2878, 1626, 1571, 1226, 1057 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 8,31 (t, 1H, 1'(amid), J = 5,5 Hz), 7,37–7,21 (m, 5H, arom.), 6,78 (d, 1H, 3, J = 8,4 Hz), 6,29 (t, 1H, 1'(urea), J = 5,5 Hz), 5,31 (d, 1H, 2, J = 8,4 Hz), 4,66, 4,64 (2t, 2H, 4', J = 5,3 Hz, J = 5,1 Hz), 3,40–3,34 (m, 4H, 3'), 3,23–2,97 (m, 4H, 2').

^{13}C NMR (DMSO-d_6, δ ppm): 171,25 (1), 157,63 (4), 141,05 (5), 128,61, 127,10 (6, 7, 9 10), 127,57 (8), 61,18, 60,11 (3'), 56,93 (2), 42,46, 42,01 (2').

1-((3-Hidroksipropilkarbamoil)(fenil)metil)-3-(3-hidroksipropil)urea (4k)

Količina reaktanta: 0,315 g spoja **3d**. Sirovi produkt **4j** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 i prekristalizacijom iz metanola i etera. Iskorištenje: 0,170 g (55 %).

t_t 182–185 ºC.

IR (KBr): ν_{max} 3289, 3092, 2942, 2883, 1626, 1561, 1059 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 8,27 (t, 1H, 1'(amid), J = 5,5 Hz), 7,36–7,22 (m, 5H, arom.), 6,65 (d, 1H, 3, J = 8,3 Hz), 6,21 (t, 1H, 1'(urea), J = 5,5 Hz), 5,26 (d, 1H, 2, J = 8,5 Hz), 4,41, 4,39 (2t, 2H, 5', J = 5,2 Hz), 3,42–3,32 (m, 4H, 4'), 3,12–2,99 (m, 4H, 2'), 1,56–1,44 (m, 4H, 3').

^{13}C NMR (DMSO-d_6, δ ppm): 171,06 (1), 157,67 (4), 141,10 (5), 128,63, 126,96 (6, 7, 9, 10), 127,59 (8), 58,79, 58,72 (4'), 56,98 (2), 39,15, 36,69 (2'), 36,33, 36,28 (3').

1-((3-Hidroksipentilkarbamoil)(fenil)metil)-3-(5-hidroksipentil)urea (4l)

Količina reaktanta: 0,315 g spoja **3d**. Sirovi produkt **4l** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 i prekristalizacijom iz metanola i etera. Iskorištenje: 0,222 g (61 %).

t_t 178–180 ºC.

IR (KBr): ν_{max} 3362, 3286, 3093, 2937, 2861, 1628, 1564, 1352, 1061 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 8,26 (t, 1H, 1'(amid), J = 5,5 Hz), 7,36–7,21 (m, 5H, arom.), 6,60 (d, 1H, 3, J = 8,4 Hz), 6,21 (t, 1H, 1'(urea), J = 5,5 Hz), 5,26 (d, 1H, 2, J = 8,4 Hz), 4,35, 4,31 (2t, 2H, 7', J = 5,3 Hz), 3,40–3,30 (m, 4H, 6'), 3,05–2,92 (m, 4H, 2'), 1,44–1,16 (m, 12H, 3'–5').

^{13}C NMR (DMSO-d_6, δ ppm): 170,90 (1), 157,49 (4), 141,21 (5), 128,59, 126,93 (6, 7, 9, 10), 127,54 (8), 61,13, 61,06 (6'), 56,94 (2), 39,63, 39,04 (2'), 32,71, 32,58 (5'), 30,31, 29,24 (3'), 23,38, 23,25 (4').

1-(1-(2-Hidroksietilkarbamoil)-2-feniletil)-3-(2-hidroksietil)urea (4m)

Količina reaktanta: 0,329 g spoja **3e**. Sirovi produkt **4m** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 i rastrljavanjem u eteru.

Iskorištenje: 0,230 g (78 %).

t_t 140–143 ºC.

IR (KBr): ν_{max} 3310, 3105, 2937, 2876, 1626, 1569, 1496, 1231, 1056 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,90 (t, 1H, 1'(amid), J = 5,1 Hz), 7,28–7,17 (m, 5H, arom.), 6,16 (d, 1H, 3, J = 8,6 Hz), 6,10 (t, 1H, 1'(urea), J = 5,1 Hz), 4,64–4,59 (m, 2H, 4), 4,37–4,30 (m, 1H, 2), 3,33–3,29 (m, 4H, 3'), 3,12–2,67 (m, 6H, 2', 5).

^{13}C NMR (DMSO-d_6, δ ppm): 172,51 (1), 158,00 (4), 138,41 (6), 129,71, 128,43 (7, 8, 10, 11), 126,58 (9), 61,16, 60,18 (3'), 54,80 (2), 42,42, 41,81 (2'), 39,42 (5).

1-(1-(3-Hidroksipropilkarbamoil)-2-feniletil)-3-(3-hidroksipropil)urea (4n)

Količina reaktanta: 0,329 g spoja **3e**. Sirovi produkt **4n** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 8,5:1,5 i rastrljavanjem u eteru.

Iskorištenje: 0,243 g (75 %).

t_t 110–113 ºC.

IR (KBr): ν_{max} 3280, 3106, 2942, 2878, 1628, 1564, 1235, 1050 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,88 (t, 1H, 1'(amid), J = 4,6 Hz), 7,27–7,14 (m, 5H, arom.), 6,04–6,02 (d, t, 2H, 3, 1'(urea)), 4,42–4,28 (m, 3H, 2, 5'), 3,39–3,34 (m, 4H, 4'), 3,12–2,69 (m, 6H, 2', 5), 1,50–1,43 (m, 4H, 3').

^{13}C NMR (DMSO-d_6, δ ppm): 172,28 (1), 158,00 (4), 138,35 (5), 129,68, 128,43 (7, 8, 10, 11), 126,58 (9), 58,93 (4'), 54,79 (2), 39,46 (5), 36,68, 36,14 (2'), 33,61, 32,72 (3').

1-(1-(5-Hidroksipentilkarbamoil)-2-feniletil)-3-(5-hidroksipentil)urea (4o)

Količina reaktanta: 0,329 g spoja **3e**. Sirovi produkt **4o** pročišćen je kromatografijom na koloni, uz pokretnu fazu diklorometan/metanol 9:1 i rastrljavanjem u eteru.

Iskorištenje: 0,197 g (52 %).

t_t 89–93 ºC.

IR (KBr): ν_{max} 3282, 3106, 2931, 2859, 1626, 1559, 1236, 1060 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,86 (t, 1H, 1'(amid), J = 5,5 Hz), 7,27–7,14 (m, 5H, arom.), 6,01 (t, 1H, 1'(urea), J = 5,5 Hz), 5,96 (d, 1H, 3, J = 8,7 Hz), 4,35–4,28 (m, 3H, 2, 7'), 3,37–3,33 (m, 4H, 6'), 3,07–2,69 (m, 6H, 2', 5), 1,43–1,16 (m, 12H, 3'–5').

^{13}C NMR (DMSO-d_6, δ ppm): 172,06 (1), 157,79 (4), 138,34 (5), 129,70, 128,40 (7, 8, 10, 11), 126,55 (9), 61,13, 61,07 (6'), 54,72 (2), 39,62, 39,55 (2'), 38,91 (5), 32,72, 32,67 (5'), 30,30, 29,33 (3'), 23,36, 23,29 (4').

2.1.6. Sinteza amida *N*-(1-benzotriazolkarbonil)aminokiselina 5a-f. Opća metoda

U otopinu odgovarajućeg klorida *N*-(1-benzotriazolkarbonil)aminokiseline **3c-e** (4 mmol) u bezvodnom toluenu (40 mL) dokapana je otopina odgovarajućeg amina (4 mmol) i TEA (0,558 mL, 4 mmol) u toluenu (30 mL). Reakcijska smjesa miješana je 1 h na sobnoj temperaturi. Istaloženi TEA hidroklorid je odsisan, a matičnica ekstrahirana vodom (3×70 mL), kloridnom kiselinom (w = 1 %, 3×70 mL) i vodom do neutralnog pH, a zatim osušena nad bezvodnim natrijevim sulfatom. Nakon uparavanja otapala pod sniženim tlakom dobiven je sirovi produkt koji kristalizira. Produkti **5a-f** prekristalizirani su iz odgovarajućeg otapala. IR spektri i tališta spojeva **5a-f** u potpunosti odgovaraju ranije sintetiziranim spojevima (Opačić *et al*, 2005).

2.1.7. Sinteza *N*- i *O*-supstituiranih hidroksiurea 6a-l. Opća metoda

U suspenziju odgovarajućeg amida *N*-(1-benzotriazolkarbonil)aminokiseline (**5a-f**) (1 mmol) i odgovarajućeg hidroksilamin hidroklorida (1,2 mmol) u 8 ml suhog toluena dodana je otopina TEA (0,167 mL, 1,2 mmol) u 8 ml suhog toluena. Reakcijska smjesa je mješana na temperaturama od sobne do 125ºC 10 minuta do 72 h. Toluen je uparen a dobivena smola je otopljena u smjesi acetona i vode te zakiseljena 10 % HCl do pH 1. Uparavanjem acetona pod sniženim tlakom taloži se sirovi produkt.

***N*-Benzhidrilamid-2-(*N*'-benziloksiureido)-L-4-metilpentanske kiseline (6a)**

Količine reaktanata: 0,442 g spoja **5a**, 0,191 g *O*-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 20 h.

Temperatura reakcije: 15,5 h na 70 ºC i 4,5 h na 114 ºC.

Sirovi produkt je pročišćen koromatografijom na koloni uz pokretnu fazu diklormetan/metanol 9,4:0,6.

Iskorištenje: 0,122 g (27 %).

t_t 100–103 ºC.

IR (KBr): ν_{max} 3308, 3061, 3030, 2957, 2930, 2872, 1645, 1525, 1496, 1453, 1336, 1240, 1212, 1078, 1028, 902, 752, 698, 607 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,18 (s, 1H, 1'), 8,82 (d, 1H, 1", J = 8,51 Hz), 7,33–7,16 (m, 15H, arom.), 6,45 (d, 1H, 3, J = 8,92 Hz), 6,03 (d, 1H, 2", J = 8,51 Hz), 4,64 (dd, 2H, 3',

$J_{vic\ H,H}$=11,6 Hz), 4,32–4,28 (m, 1H, 2), 1,43–1,32 (m, 3H, 5, 6), 0,79 (2d, 6H, 7, 8, J = 6,43 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 171,66 (1), 158,95 (4), 142,25, 142,21 (3", 9"), 136,26 (4'), 128,73, 128,35, 128,30, 128,26, 127,39, 127,09 (5', 6', 8', 9', 4", 5", 7", 8", 10", 11", 13", 14"), 128,09, 126,94, 126,90 (7', 6", 12"), 77,40 (3'), 55,84 (2"), 51,16 (2), 41,67 (5), 24,17, 23,04, 21,72 (6–8).

_N_-Benzhidrilamid-2-(_N_'-metil-_N_'-hidroksiureido)-L-4-metilpentanske kiseline (6b)

Količine reaktanta: 0,442 g spoja **5a**, 0,100 g _N_-metilhidroksilamin hidroklorida.

Trajanje reakcije: 0,15 h.

Temperatura: 125 ºC.

Reakcija rađena u talini bez otapala. Sirovi produkt je rastrljan u eteru.

Iskorištenje: 0,283 g (77 %).

t_t 219–222 ºC (raspad).

IR (KBr): ν_{max} 3407, 3309, 3162, 3031, 2964, 2930, 2900, 2872, 1648, 1548, 1515, 1496, 1471, 1450, 1437, 1368, 1296, 1185, 1173, 1120, 761, 749, 701 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,52 (s, 1H, 2'), 8,90 (d, 1H, 1", J = 8,47 Hz), 7,38–7,18 (m, 10H, arom.), 6,68 (d, 1H, 3, J = 8,97 Hz), 6,10 (d, 1H, 2", J = 8,47 Hz), 4,38–4,32 (m, 1H, 2), 2,95 (s, 3H, 3'), 1,58–1,39 (m, 3H, 5, 6), 0,85 (2d, 6H, 7, 8, J = 3,24 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 171,81 (1), 160,34 (4), 142,30 (3", 9"), 128,39, 128,35, 127,41, 127,10 (4", 5", 7", 8", 10", 11", 13", 14"), 127,01, 126,97 (6", 12"), 55,84 (2"), 51,85 (2), 42,03 (5), 38,59 (3'), 24,24, 23,05, 21,84 (6–8).

_N_-Cikloheksanmetilamid-2-(_N_'-benziloksiureido)-D-2-feniletanske kiseline (6c)

Količine reaktanata: 0,391 g spoja **5b**, 0,191 g _O_-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 5 h.

Temperatura reakcije: 114 ºC.

Sirovi produkt je pročišćen koromatografijom na koloni uz pokretnu fazu cikloheksan/etil--acetat/metanol 3:1:0,6.

Iskorištenje: 0,117 g (30 %).

t_t 148–150 ºC.

IR (KBr): ν_{max} 3310, 3091, 3063, 3032, 2926, 2854, 1639, 1533, 1450, 1355, 1221, 1154, 985, 931, 907, 747, 697 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,40 (s, 1H, 1'), 8,29 (t, 1H, 1'', J = 5,57 Hz), 7,43–7,22 (m, 10H, arom.), 6,91 (d, 1H, 3, J = 7,98 Hz), 5,28 (d, 1H, 2, J = 7,98 Hz), 4,76 (dd, 2H, 3', $J_{\text{vic H,H}}$=11,26 Hz), 2,93–2,82 (m, 2H, 2''), 1,61–0,83 (m, 11H, 3''–8'').

^{13}C NMR (DMSO-d_6, δ ppm): 169,68 (1), 158,24 (4), 139,79 (5), 136,09 (4'), 129,00, 128,42, 128,20, 126,37 (6, 7, 9, 10, 5', 6', 8', 9'), 128,29, 127,36 (8, 7'), 77,54 (3'), 55,76 (2), 44,88 (2''), 37,35 (3''), 30,19, 30,14, 25,32 (4'', 5'', 7'', 8''), 25,93 (6'').

_N_-Cikloheksanmetilamid-2-(_N_'-metil-_N_'-hidroksiureido)-D-2-feniletanske kiseline (6d)

Količine reaktanata: 0,391 g spoja **5b**, 0,100 g _N_-metilhidroksilamin hidroklorida.

Trajanje reakcije: 24 h.

Temperatura reakcije: s.t.

Sirovi produkt je pročišćen koromatografijom na koloni uz pokretnu fazu cikloheksan/etil--acetat 1:1.

Iskorištenje: 0,127 g (40 %).

t_t 123–126 ºC.

IR (KBr): ν_{max} 3425, 3310, 2925, 2853, 1650, 1532, 1450, 1367, 1184, 1120, 698, 668 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,85 (bs, 1H, 2'), 8,33 (t, 1H, 1'', J = 5,76 Hz), 7,39–7,23 (m, 5H, arom.), 7,07 (d, 1H, 3, J = 8,16 Hz), 5,28 (d, 1H, 2, J = 8,16 Hz), 2,95 (s, 3H, 3'), 2,92–2,87 (m, 2H, 2''), 1,60–0,77 (m, 11H, 3''–8'').

^{13}C NMR (DMSO-d_6, δ ppm): 170,32 (1), 160,05 (4), 140,51 (5), 128,69, 126,89 (6, 7, 9, 10), 127,84 (8), 56,86 (2), 45,34 (2''), 38,76 (3'), 37,81 (3''), 30,66, 30,62, 25,78 (4'', 5'', 7'', 8''), 26,39 (6'').

_N_-Benzhidrilamid-2-(_N_'-benziloksiureido)-D-2-feniletanske kiseline (6e)

Količine reaktanata: 0,462 g spoja **5c**, 0,191 g _O_-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 4,5 h.

Temperatura reakcije: 114 ºC.

Sirovi produkt je prekristaliziran iz acetona i vode.

Iskorištenje: 0,093 g (20 %).

t_t 161–165 ºC.

IR (KBr): ν_{max} 3308, 3062, 3031, 2934, 1667, 1642, 1536, 1496, 1453, 1366, 1231, 1213, 1031, 934, 743, 700, 640, 614 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,33 (s, 1H, 1'), 9,19 (d, 1H, 1", J = 8,28 Hz), 7,35–6,98 (m, 20H, arom.), 6,85 (d, 1H, 3, J = 7,96 Hz), 6,01 (d, 1H, 2", J = 7,96 Hz), 5,43 (d, 1H, 2, J = 7,96 Hz), 4,68 (dd, 2H, 3', $J_{\text{vic H,H}}$=10,94 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 169,08 (1), 158,25 (4), 141,90, 141,77 (3", 9"), 139,34 (5), 136,05 (4'), 128,94, 128,42, 128,38, 128,23, 127,51, 126,79, 126,46 (6, 7, 9, 10, 4', 5', 7', 8', 4", 5", 7", 8", 10", 11", 13", 14"), 77,52 (3'), 56,02, 55,63 (2, 2").

_N_-Benzhidrilamid-2-(_N_'-metil-_N_'-hidroksiureido)-D-2-feniletanske kiseline (6f)

Količine reaktanata: 0,462 g spoja **5c**, 0,100 g _N_-metilhidroksilamin hidroklorida.

Trajanje reakcije: 1,75 h.

Temperatura reakcije: 70 ºC.

Iskorištenje: 0,331 g (85 %).

t_t 165–168 ºC (raspad).

IR (KBr): ν_{max} 3439, 3376, 3210, 3064, 3031, 2924, 2892, 1637, 1584, 1520, 1496, 1450, 1367, 1177, 1120, 1030, 748, 697, 636 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,71 (s, 1H, 2'), 9,31 (d, 1H, 1", J = 8,40 Hz), 7,45–7,04 (m, 16H, arom., 3), 6,09 (d, 1H, 2", J = 8,19 Hz), 5,53 (d, 1H, 2, J = 8,19 Hz), 2,96 (s, 3H, 3').

^{13}C NMR (DMSO-d_6, δ ppm): 169,72 (1), 160,03 (4), 142,47, 142,25 (3", 9"), 140,11 (5), 128,93, 128,77, 128,74, 127,97, 127,31, 127,01 (6, 7, 9, 10, 4", 5", 7", 8", 10", 11", 13", 14"), 27,97, 127,66, 127,43 (8, 6", 12"), 56,68, 56,51 (2, 2"), 38,78 (3').

_N_-Cikloheksilamid-2-(_N_'-benziloksiureido)-L-3-fenilpropanske kiseline (6g)

Količine reaktanata: 0,391 g spoja **5d**, 0,191 g _O_-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 1 h.

Temperatura reakcije: 114 ºC.

Sirovi produkt je pročišćen rastrljavanjem u eteru.

Iskorištenje: 0,267 g (65 %).

t_t 129–131 ºC.

IR (KBr): ν_{max} 3296, 3206, 3064, 3031, 2931, 2854, 1639, 1533, 1498, 1453, 1364, 1250, 1075, 952, 814, 750, 698 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,24 (s, 1H, 1'), 7,88 (d, 1H, 1", J = 7,66 Hz), 7,35–7,13 (m, 10H, arom.), 6,53 (d, 1H, 3, J = 8,61 Hz), 4,64 (s, 1H, 3'), 4,41 (q, 1H, 2, J = 7,48 Hz), 3,52–3,49 (m, 1H, 2"), 2,96–2,88 (d, 2H, 5), 1,73–1,03 (m, 10H, 3"–7").

^{13}C NMR (DMSO-d_6, δ ppm): 170,36 (1), 159,14 (4), 137,93 (6), 136,59 (4'), 129,82, 129,17, 128,36 (7, 8, 10, 11, 5', 6', 8', 9'), 128,60, 126,63 (9, 7'), 77,84 (3'), 54,00 (2), 47,96 (2''), 39,01 (5), 32,74, 32,73, 24,90, 24,85 (3'', 4'', 6'', 7''), 25,65 (5'').

_N_-Cikloheksilamid-2-(_N_'-metil-_N_'-hidroksiureido)-L-3-fenilpropanske kiseline (6h)

Količine reaktanata: 0,391 g spoja **5d**, 0,100 g _N_-metilhidroksilamin hidroklorida.

Trajanje reakcije: 48 h.

Temperatura reakcije: s.t.

Iskorištenje: 0,183 g (57 %).

t_t 153–155 ºC.

IR (KBr): ν_{max} 3390, 3289, 3089, 2935, 2854, 1648, 1627, 1550, 1452, 1390, 1182, 751, 702 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,50 (s, 1H, 2'), 7,83 (d, 1H, 1'', J = 7,84 Hz), 7,27–7,15 (m, 5H, arom.), 6,59 (d, 1H, 3, J = 8,56 Hz), 4,36 (q, 1H, 2, J = 6,82 Hz, 8,27 Hz), 3,52–3,43 (m, 1H, 2''), 2,91 (s, 3H, 3'), 2,88 (d, 2H, 5, J = 6,97 Hz), 1,71–1,02 (m, 10H, 3''–7'').

^{13}C NMR (DMSO-d_6, δ ppm): 170,38 (1), 160,61 (4), 137,91 (6), 129,77, 128,43 (8, 7, 10, 11), 126,68 (9), 54,62 (2), 47,86 (2''), 39,32 (5), 38,99 (3'), 32,70, 24,92, 24,87 (3'', 4'', 6'', 7''), 25,64 (5'').

_N_-Cikolheksanmetilamid-2-(_N_'-benziloksiureido)-L-3-fenilpropanske kiseline (6i)

Količine reaktanata: 0,405 g spoja **5e**, 0,191 g _O_-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 4,5 h.

Temperatura reakcije: 70 ºC.

Sirovi produkt je pročišćen rastrljavanjem u eteru.

Iskorištenje: 0,243 g (59 %).

t_t 130–131 ºC.

IR (KBr): ν_{max} 3313, 3214, 3064, 3031, 2921, 2852, 1661, 1640, 1542, 1496, 1449, 1363, 1276, 1242, 1072, 948, 813, 748, 698, 632, 616 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,24 (s, 1H, 1'), 7,96 (t, 1H, 1'', J = 5,57 Hz), 7,39–7,19 (m, 10H, arom.), 6,56 (d, 1H, 3, J = 8,44 Hz), 4,65 (s, 1H, 3'), 4,43 (q, 1H, 2, J = 8,12, 5,89 Hz), 2,99–2,82 (m, 4H, 2'', 5), 1,66–0,81 (m, 11H, 3''–8'').

^{13}C NMR (DMSO-d_6, δ ppm): 170,38 (1), 158,12 (4), 136,98 (4'), 135,56 (6), 128,68, 128,08, 127,69, 127,43 (7, 8, 10, 11, 5', 6'. 8', 9'), 127,52, 125,67 (9, 7'), 76,78 (3'), 53,22 (2), 44,24 (2''), 37,81 (5), 36,74 (3''), 29,72, 24,82 (4'', 5'', 7'', 8''), 25,42 (6'').

_N_-Cikolheksanmetilamid-2-(_N_'-metil-_N_'-hidroksiureido)-L-3-fenilpropanske kiseline(6j)

Količine reaktanata: 0,405 g spoja **5e**, 0,100 g _N_-metilhidroksilamin hidroklorida.

Trajanje reakcije: 2 h.

Temperatura reakcije: 40 ºC.

Sirovi produkt je pročišćen rastrljavanjem u eteru.

Iskorištenje: 0,268 g (80 %).

t_t 157–159 ºC.

IR (KBr): ν_{max} 3371, 3329, 3194, 2924, 2853, 1658, 1642, 1580, 1528, 1495, 1445, 1190, 1123, 746, 700, 669 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,48 (s, 1H, 2'), 7,93 (t, 1H, 1'', J = 5,21 Hz), 7,29–7,16 (m, 5H, arom.), 6,63 (d, 1H, 3, J = 8,54 Hz), 4,38 (q, 1H, 2, J = 7,32 Hz), 2,97–2,75 (m, 7H, 5, 2''), 2,91 (s, 3H, 3'), 1,69–0,76 (m, 11H, 3''–8'').

^{13}C NMR (DMSO-d_6, δ ppm): 171,48 (1), 160,64 (4), 138,03 (6), 129,68, 128,47 (7, 8, 10, 11), 126,67 (9), 54,88 (2), 45,26 (2''), 39,14 (5), 38,98 (3'), 37,77 (3''), 30,76, 25,87 (4'', 5'', 7'', 8''), 26,46 (6'').

_N_-Benzhidrilamid-2-(_N_'-benziloksiureido)-L-3-fenilpropanske kiseline (6k)

Količine reaktanata: 0,476 g spoja **5f**, 0,191 g _O_-benzilhidroksilamin hidroklorida.

Trajanje reakcije: 52,5 h.

Temperatura: 70 ºC.

Sirovi produkt je pročišćen rastrljavanjem u eteru.

Iskorištenje: 0,185 g (39 %).

t_t 161–164 ºC.

IR (KBr): ν_{max} 3288, 3062, 3030, 1649, 1535, 1495, 1454, 1229, 1030, 910, 750, 698 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,17 (s, 1H, 1'), 8,89 (d, 1H, 1'', J = 8,47 Hz), 7,32–7,11 (m, 20H, arom.), 6,54 (d, 1H, 3, J = 8,74 Hz), 6,03 (d, 1H, 2'', J = 8,34 Hz), 4,54 (q+s, 3H, 2, 3''), 2,93–2,84 (m, 2H, 5).

^{13}C NMR (DMSO-d_6, δ ppm): 170,55 (1), 158,75 (4), 142,15, 142,14 (3'', 9''), 137,41 (6), 136,10 (4'), 129,31, 128,62, 128,34, 128,25, 128,00, 127,39, 127,14 (7, 8, 10, 11, 4'', 5'', 7'',

8", 10", 11", 13", 14"), 128,07, 127,00, 126,91, 126,28 (9, 6", 12"), 77,34 (5), 55,96, 53,75 (2, 2"), 38,16 (3').

***N*-Benzhidrilamid-2-(*N*'-metil-*N*'-hidroksiureido)-L-3-fenilpropanske kiseline (6l)**

Količine reaktanata: 0,476 g spoja **5f**, 0,100 g *N*-metilhidroksilamin hidroklorida.

Temperatura: s.t.

Trajanje reakcije: 72 h.

Sirovi produkt je pročišćen rastrljavanjem u eteru.

Iskorištenje: 0,179 g (44 %).

t_t 152–154 ºC (raspad).

IR (KBr): ν_{max} 3432, 3347, 3062, 3030, 2867, 1664, 1629, 1536, 1496, 1450, 1384, 1230, 1179, 1117, 1030, 744, 700 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, *J*/Hz): 9,52 (s, 1H, 2'), 8,96 (d, 1H, 1", *J* = 8,32 Hz), 7,37–7,15 (m, 15H, arom.), 6,69 (d, 1H, 3, *J* = 8,32 Hz), 6,09 (d, 1H, 2", *J* = 8,32 Hz), 4,60 (q, 1H, 2, *J* = 6,93 Hz), 2,95–2,91 (m, 2H, 5), 2,91 (s, 3H, 3').

^{13}C NMR (DMSO-d_6, δ ppm): 171,10 (1), 160,71 (4), 142,70, 142,63 (3", 9"), 137,91 (6), 129,78, 128,84, 128,78, 128,51, 127,89, 127,61 (7, 8, 10, 11, 4", 5", 7", 8", 10", 11", 13", 14"), 127,51, 127,39, 126,73 (9, 6", 12"), 56,37, 54,87 (2, 2"), 38,98 (3').

2.1.8. Sinteza 1-karbamoilbenzotriazola (amida 1-benzotriazolkarboksilne kiseline) 7a-g

Metoda A. U otopinu BtcCl (**1**) (1,810 g, 10 mmol) u bezvodnom toluenu dokapana je otopina TEA (1,394mL, 10 mmol) i odgovarajućeg amina (10 mmol) u toluenu tijekom 0,25 h. Nakon dokapavanja reakcijska smjesa miješana je 1 h na sobnoj temperaturi. Reakcijska smjesa uparena je pod sniženim tlakom, zatim je otopljena u smjesi etil-acetata i vode (1:1, 30 mL). Organski sloj ekstrahiran je vodom (3×30 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Dobiveni sirovi 1-karbamoilbenzotriazoli **7a-f** pročišćeni su prekristalizacijom iz etera/petroletera (**7a**), metanola i vode (**7b-d**, **7f**) ili acetona i vode (**7e**). IR spektri i tališta spojeva **7b** i **7e** u potpunosti odgovaraju ranije sintetiziranim spojevima (Butula *et al*, 1977, Butula *et al*, 1978).

1-(*N*-Ciklopentilkarbamoil)benzotriazol (7a)

Metoda A. Količina reaktanta: 0,852 g ciklopentilamina.

Iskorištenje: 1,428 g (62 %).

t_t 67–68 ºC.

IR (KBr): ν_{max} 3255, 2949, 2866, 1736, 1523, 1487, 1448, 1371, 1287, 1233, 1185, 1130, 1073, 1000, 944, 925, 815, 747, 652 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, *J*/Hz): 9,09 (d, 1H, 1', *J* = 6,62 Hz), 8,19 (d, 2H, arom., *J* = 8,27 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,72 Hz, *J* = 6,62 Hz), 4,29–4,22 (m, 1H, 2'), 1,97–1,56 (m, 8H, 3'–6').

^{13}C NMR (DMSO-d_6, δ ppm): 148,98 (1), 145,87, 131,85 (2, 7), 130,24, 125,90, 120,16, 114,11 (3–6), 52,55 (2'), 32,18, 24,01 (3'–6').

1-(*N*-Cikloheksilkarbamoil)benzotriazol (7b)

Metoda A. Količina reaktanta: 0,992 g cikloheksilamina.

Iskorištenje: 1,710 g (70 %).

t_t 73–74 °C (Butula *et al*, 1977).

IR (KBr): ν_{max} 3327, 2938, 2856, 1733, 1517, 1488, 1447, 1360, 1285, 1226, 1151, 1090, 1059, 1002, 968, 928, 835, 791, 755, 612 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, *J*/Hz): 8,98 (d, 1H, 1', *J* = 8,18 Hz), 8,19 (d, 2H, arom., *J* = 9,20 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,67 Hz), 3,81–3,73 (m, 1H, 2'), 1,93–1,11 (m, 10H, 3'–7').

^{13}C NMR (DMSO-d_6, δ ppm): 148,61 (1), 145,91, 131,89 (2, 7), 130,24, 125,94, 120,17, 114,05 (3–6), 50,34 (2'), 32,41, 25,35 (3', 4', 6', 7'), 25,46 (5').

1-(*N*-Cikloheksanmetilkarbamoil)benzotriazol (7c)

Metoda A. Količina reaktanta: 1,132 g cikloheksanmetilamina.

Iskorištenje: 2,531 g (98 %).

t_t 66–68 °C.

IR (KBr): ν_{max} 3360, 2925, 2852, 1739, 1527, 1488, 1448, 1364, 1286, 1232, 1152, 1072, 1011, 954, 786, 771, 753, 593 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, *J*/Hz): 9,18 (t, 1H, 1', *J* = 5,62 Hz), 8,20 (d, 2H, arom., *J* = 9,20 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,67 Hz), 3,23 (t, 2H, 2', *J* = 6,14 Hz), 1,77–1,58, 1,27–0,93 (2m, 11H, 3'–8').

^{13}C NMR (DMSO-d_6, δ ppm): 149,54 (1), 145,94, 131,83 (2, 7), 130,28, 125,90, 120,18, 114,05 (3–6), 40,60 (2'), 37,77 (3'), 30,76, 25,78 (4', 5', 7', 8'), 25,86 (6').

1-(*N*-Benzilkarbamoil)benzotriazol (7d)

Metoda A. Količina reaktanta: 1,072 g benzilamina.

Iskorištenje: 1,867 g (74 %).

t_t 110 °C.

IR (KBr): ν_{max} 3362, 1733, 1526, 1486, 1447, 1358, 1288, 1256, 1231, 1130, 1076, 1002, 793, 752, 696, 609, 580, 562 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,78 (t, 1H, 1', J = 5,81 Hz), 8,20 (d, 2H, arom., J = 8,72 Hz), 7,72, 7,55 (2t, 2H, arom., J = 7,89 Hz), 7,45–7,25 (m, arom., 5H), 4,59 (d, 2H, 2', J = 6,23 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 149,67 (1), 145,96, 131,84 (2, 7), 139,10 (3'), 130,39, 125,98, 120,24, 114,02 (3–6), 128,84, 127,88 (4', 5', 7', 8'), 127,54 (6'), 43,94 (2').

1-(*N*-Feniletilkarbamoil)benzotriazol (7e)

Metoda A. Količina reaktanta: 1,212 g feniletilamina.

Iskorištenje: 2,157 g (74 %).

t_t 114–115 °C (lit. 115–117 °C, Butula *et al*, 1978).

IR (KBr): ν_{max} 3369, 3024, 2949, 1736, 1530, 1497, 1447, 1363, 1288, 1231, 1128, 1097, 1034, 1002, 920, 840, 749, 699, 588, 563 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,27 (t, 1H, 1', J = 6,06 Hz), 8,21 (d, 2H, arom., J = 8,48 Hz), 7,71, 7,54 (2t, 2H, arom., J = 7,27 Hz), 7,34–7,19 (m, arom., 5H), 3,63 (q, 2H, 2', J = 6,67 Hz), 2,98 (t, 2H, 3', J = 7,88 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 149,37 (1), 145,92, 131,78 (2, 7), 139,42 (4'), 130,34, 125,94, 120,20, 114,01 (3–6), 129,15, 128,85 (5', 6', 8', 9'), 126,69 (7'), 41,96, 35,36 (2', 3').

1-(*N*-Benzhidrilkarbamoil)benzotriazol (7f)

Metoda A. Količina reaktanta: 1,833 g benzhidrilamina.

Iskorištenje: 2,135 g (65 %).

t_t 119–122 °C.

IR (KBr): ν_{max} 3288, 3028, 2931, 1707, 1520, 1449, 1375, 1293, 1231, 1069, 1031, 931, 864, 752, 695, 644, 607, 535 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 10,00 (d, 1H, 1', J = 8,57 Hz), 8,21, 8,17 (2d, 2H, arom., J = 8,04 Hz), 7,74–7,28 (m, 12H, arom.), 6,40 (d, 1H, 2', J = 9,11 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 149,16 (1), 145,93, 131,95 (2, 7), 141,62 (3', 9'), 130,42, 126,08, 120,27, 113,96 (3–6), 128,87, 128,23 (4', 5', 7', 8', 10', 11', 13', 14'), 127,82 (6', 12'), 58,37 (2').

1-(*N*-Benziloksikarbamoil)benzotriazol (7g)

Metoda B. Otopina BtcCl (**1**) (1,810 g, 10 mmol) u bezvodnom dioksanu dokapana je u suspenziju *O*-benzilhidroksilamin hidroklorida (1,596 g, 10 mmol) i TEA (2,788 mL, 20 mmol) u dioksanu tijekom 0,25 h. Nakon dokapavanja reakcijska smjesa miješana je 2 h na sobnoj temperaturi. Talog TEA hidroklorida je odsisan a matičnica je uparena pod sniženim tlakom. Dobiven sirovi produkt **7g** je prekristailziran iz smjese etil-acetat/eter. IR spektar i talište spoja **7g** u potpunosti odgovara ranije sintetiziranom spoju (Butula *et al*, 2000).

Iskorištenje: 2,254 g (84 %).

t_t 113–114 °C (Butula *et al*, 2000).

IR (KBr): ν_{max} 3246, 3133, 3030, 2947, 2884, 1728, 1603, 1484, 1367, 1289, 1231, 1132, 1093, 1003, 924, 869, 751, 699, 563 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,95 (s, 1H, 1'), 8,27, 8,09 (2d, 2H, J = 8,20 Hz, 8,50 Hz), 7,67, (t, 2H, arom., J = 7,7 Hz), 7,48–7,34 (m, arom., 5H), 5,14 (s, 2H, 2').

^{13}C NMR (DMSO-d_6, δ ppm): 148,68 (1), 145,72, 131,54 (2, 7), 134,46 (3'), 130,23, 125,66, 120,07, 113,49 (3–6), 129,28, 128,63 (4', 5', 7', 8'), 126,97 (6'), 79,24 (2').

2.1.9. Sinteza 4-benziloksisemikarbazida (8)

U otopinu hidrazin hidrata (0,534 mL, 11 mmol) u dioksanu dokapana je otopina 1-(*N*-benziloksikarbamoil)benzotriazola (**7g**) (2,683 g, 10 mmol) u dioksanu tijekom 0,25 h. Reakcijska smjesa je miješana 1 h na s.t. i zatim uparena. Sirovi produkt **8** kromatografiran je na koloni, uz pokretnu fazu diklorometan/metanol 9,5:0,5 te je rastrljan u eteru.

Iskorištenje: 1,486 g (82 %).

t_t 90–92 °C.

IR (KBr): ν_{max} 3332, 3287, 3217, 3061, 3030, 2962, 2925, 2875, 1655, 1637, 1517, 1456, 1366, 1325, 1228, 1210, 1192, 1096, 1070, 980, 915, 797, 751, 702, 680, 607 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,18 (s, 1H, 4), 7,84 (s, 1H, 2), 7,42–7,29 (m, 5H, arom.), 4,71 (s, 2H, 5), 4,01 (s, 2H, 1).
^{13}C NMR (DMSO-d_6, δ ppm): 161,67 (3), 137,18 (6), 129,08, 128,63 (7, 8, 10, 11), 128,37 (9), 77,70 (5).

2.1.10. Sinteza 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazida (9)

U otopinu BtcCl (**1**) (1,357 g, 8 mmol) u bezvodnom dioksanu dokapana je otopina 4-benziloksisemikarbazida **8** (1,450 g, 8 mmol) i TEA (1,115 mL, 8 mmol) u bezvodnom dioksanu tijekom 0,25 h. Reakcijska smjesa miješana je na sobnoj temperaturi 1 h, zatim uparena je pod sniženim tlakom te otopljena u smjesi etil-acetata i vode (1:1, 40 mL). Organski sloj ekstrahiran je vodom (3×40 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Sirovi produkt **9** pročišćen je prekristalizacijom iz etera.
Iskorištenje: 1,906 g (73 %).
t_t 90–93 °C (raspad).
IR (KBr): ν_{max} 3482, 3376, 3340, 3164, 3005, 2924, 2854, 1738, 1674, 1556, 1487, 1449, 1387, 1287, 1235, 1066, 1033, 905, 754, 744, 704, 680, 652, 609, 580, 539 cm^{-1}.
^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 10,97, 9,87, 9,31 (3s, 3H, 1, 2, 4), 8,26–8,18 (2d, 2H, arom.), 7,77, 7,59 (2t, 2H, arom.), 7,49–7,35 (m, 5H, arom., 7–11), 4,85 (s, 2H, 5).
^{13}C NMR (DMSO-d_6, δ ppm): 159,37 (3), 149,86 (1'), 145,66, 131,92 (2', 7'), 136,74 (6), 130,78, 126,31, 120,42, 113,79 (3'–6'), 129,20, 128,68 (7, 8, 10, 11), 128,54 (9), 78,11 (5).

2.1.11. Sinteza NSAID benzotriazolida 10a-d. Opća metoda

U otopinu odgovarajućeg NSAID (ibuprofena, fenoprofena, ketoprofena i reduciranog derivata ketoprofena) (10 mmol) i TEA (1,393 mL, 10 mmol) u bezvodnom toluenu (30 mL) dokapana je otopina BtcCl (**1**) (1,696 g, 10 mmol) u bezvodnom toluenu (30 mL) tijekom 0,25 h. Reakcijska smjesa miješana je na sobnoj temperaturi 2 h, a zatim ekstrahirana vodom (3×60 mL). Organski sloj sušen je nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Dobiveni sirovi benzotriazolid je zatim prekristaliziran iz smjese acetona i vode (**10a**,**b**) ili rastrljan eterom (**10c**,**d**). Tališta spojeva **10a-d** i njihovi

spektroskopski podaci u potpunosti odgovaraju ranije sintetiziranim spojevima (Rajić *et al*, 2010; Zorc *et al*, 1994; Zorc *et al*,1993).

2.1.12. Sinteza hidrazida NSAID 11a-c. Opća metoda

Otopina odgovarajućeg NSAID benzotriazolida **10a-c** (10 mmol) u dioksanu dokapana je otopini hidrazin hidrata (50 mmol) u dioksanu tijekom 0,25 h. Reakcijska smjesa miješana je na sobnoj temperaturi 1 h, uparena pod sniženim tlakom i otopljena u etil-acetatu (30 mL). Organski sloj ekstrahiran je zaluženom vodom (3×30 mL, 10 kapi 5 % NaOH) i vodom (1×30 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Produkti **11a-c** prekristalizirani su iz etera/petroletera (**11a**,**c**) ili pročišćeni kromatografijom na koloni (**11b**).

2-(4-Izobutilfenil)propanhidrazid (11a)

Količina reaktanta: 3,074 g spoja **10a**. Sirovi produkt **11a** pročišćen je prekristalizacijom iz etera/petroletera.

Iskorištenje: 1,96 g (89 %).

t_t 80 ºC (lit. 73–75 ºC, Sharma *et al*, 2003).

IR (KBr): ν_{max} 3276, 3180, 3052, 2954, 2919, 1688, 1641, 1535, 1510, 1452, 1416, 1358, 1257, 1237, 1124, 1074, 1007, 965, 848, 800, 786, 731, 690, 521 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,12 (s, 1H, 2), 7,12 (dd, 4H, arom., J = 8,32 Hz, 38,27 Hz), 4,15 (s, 2H, 1), 3,46 (q, 1H, 4, J = 6,96 Hz), 2,38 (d, 2H, 12, J = 6,96 Hz), 1,82–1,73 (m, 1H, 13), 1,30 (d, 3H, 5, J = 6,96 Hz), 0,83 (d, 6H, 14,15, J = 6,65 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,37 (3), 139,78, 139,70 (6, 9), 129,15, 127,44 (7, 8, 10, 11), 44,70 (12), 43,37 (4), 30,09 (13), 22,63 (14, 15), 18,82 (5).

2-(3-Fenoksifenil)propanhidrazid (11b)

Količina reaktanta: 3,434 g spoja **10b**. Sirovi produkt **11b** pročišćen je kromatografijom na koloni uz pokretnu fazu diklormetan/metanol 9,5:0,5.

Iskorištenje: 2,307 g (90 %).

Ulje.

IR (KBr): ν_{max} 3285, 3040, 2978, 1659, 1631, 1582, 1487, 1445, 1244, 1210, 1164, 963, 928, 757, 692 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,19 (s, 1H, 2), 7,43–6,82 (m, 9H, arom.), 4,25 (s, 2H, 1), 3,53 (q, 1H, 4, J = 6,99 Hz), 1,32 (d, 3H, 5, J = 7,14 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,87 (3), 157,01, 156,92, 144,73 (6, 10, 12), 130,50, 119,04 (13, 14, 16, 17), 130,15, 123,88, 122,86, 118,06, 117,02 (7–9, 11, 15), 43,58 (4), 18,76 (5).

2-(3-Benzilfenil)propanhidrazid (11c)

Količina reaktanta: 3,554 g spoja **10c**. Sirovi produkt **11c** pročišćen je prekristalizacijom iz etera/petroletera.

Iskorištenje: 2,0 g (78 %).

t_t 80–82 ºC.

IR (KBr): ν_{max} 3289, 3191, 3168, 3063, 3023, 2975, 2919, 1637, 1601, 1534, 1486, 1453, 1382, 1261, 1008, 976, 786, 759, 722, 696, 685, 599 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,14 (s, 1H, 2), 7,31–7,03 (m, 9H, arom.), 4,17 (s, 2H, 1), 3,90 (s, 2H, 12), 3,48 (q, 1H, 4, J = 6,91 Hz), 1,33 (d, 3H, 5, J = 7,27 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,19 (3), 142,66, 141,65, 141,47 (6, 10, 13), 129,14, 128,88 (13, 14, 16, 17), 128,69, 128,15, 127,38, 126,42, 125,44 (7–9, 11, 15), 43,72 (4), 41,64 (12), 18,88 (5).

2.1.13. Sinteza estera ketoprofena i ureidoamida 12a-d. Opća metoda

Benzotriazolid ketoprofena (**10d**) (0,341 g, 1 mmol), odgovarajući ureidoamid **4b**,**i**,**j**,**m** (0,5 mmol) i TEA (0,279 mL, 2 mmol) pomiješani su i zagrijani na temperaturu taljenja reakcijske smjese. Reakcijska smjesa je miješana na toj temperaturi 0,25 h, ohlađena i otopljena u etil-acetatu (30 mL). Organski sloj ekstrahiran je zaluženom vodom (3×30 mL, 10 kapi 5 % NaOH) i vodom (1×30 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Produkti **12a-d** su pročišćeni kromatografijom na koloni uz pokretnu fazu diklormetan/metanol 9,5:0,5 (**12a**,**b**) ili etil-acetat/petroleter/metanol 3:1:0,1 (**12c**,**d**).

15-(3-Benzoilfenil)-6-metil-5,8,14-triokso-13-oksa-4,7,9-triazaheksadecil-2-(3-benzoil-fenil)propanoat (**12a**)

Količina reaktanta: 0,124 g spoja **4b**.

Temperatura reakcije: 125 °C.

Iskorištenje: 0,295 g (82 %).

Ulje.

IR (KBr): ν_{max} 3302, 3065, 2927, 2869, 1731, 1659, 1635, 1569, 1556, 1449, 1380, 1318, 1284, 1250, 1206, 1173, 1079, 960, 719, 644 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,91 (t, 1H, 1'(amid), J = 5,62 Hz), 7,74–7,50 (m, 18H, arom.), 6,08 (t, 1H, 1'(urea), J = 5,62 Hz), 6,01 (d, 1H, 3, J = 7,79 Hz), 4,10–3,90 (m, 7H, 2, 4', 2''), 3,08–2,93 (m, 4H, 2'), 1,70–1,59 (m, 4H, 3'), 1,43 (d, 6H, 3'', J = 7,07 Hz), 1,11 (d, 3H, 5, J = 6,89 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 196,07 (10''), 174,00, 173,95 (1''), 173,60 (1), 157,72 (4), 141,58, 137,65, 137,41 (4'', 8'', 11''), 133,20, 132,22, 129,30, 128,95, 128,90, (5''–7'', 9'', 14''), 130,05, 129,04 (12'', 13'', 15'', 16''), 62,82, 62,73 (4'), 49,01 (2), 44,71 (2''), 36,23, 35,57 (2'), 29,61, 28,70 (3'), 20,19 (5), 18,91 (3'').

2-(3-Benzoilfenil)-12-izobutil-3,11,14-triokso-4-oksa-10,13,15-triazaikozan-20-il-2-(3--benzoilfenil)propanoat (**12b**)

Količina reaktanta: 0,173 g spoja **4i**.

Temperatura reakcije: 100 °C.

Iskorištenje: 0,299 g (73 %).

Ulje.

IR (KBr): ν_{max} 3291, 3064, 2932, 2869, 1732, 1659, 1633, 1566, 1556, 1448, 1378, 1318, 1283, 1206, 1176, 1077, 954, 721, 706, 643 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 7,87 (t, 1H, 1'(amid), J = 5,50 Hz), 7,73–7,50 (m, 18H, arom.), 5,92 (t, 1H, 1'(urea), J = 5,50 Hz), 5,87 (d, 1H, 3, J = 8,84 Hz), 4,13–3,89 (m, 7H, 2, 6', 2''), 3,02–2,86 (m, 4H, 2'), 1,54–1,13 (m, 21H, 5, 6, 3'–5', 3''), 0,84 (2d, 6H, 7, 8, J = 5,85 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 196,04 (10''), 173,95 (1''), 173,30 (1), 157,95 (4), 141,62, 137,63, 137,41 (4'', 8'', 11''), 133,21, 132,20, 129,36, 128,92, (5''–7'', 9'', 14''), 130,03, 129,04 (12'', 13'', 15'', 16''), 64,75 (6'), 51,89 (2), 44,75 (2''), 43,00 (5), 39,43, 38,56 (2'), 29,95, 28,95, 28,18, 28,09, 23,05, 23,00 (3'–5'), 24,73, 23,38, 22,54 (6–8), 18,82 (3'').

13-(3-Benzoilfenil)-4,7,12-triokso-5-fenil-11-oksa-3,6,8-triazatetradecil 2-(3-benzoil-fenil)propanoat (**12c**)

Količina reaktanta: 0,141 g spoja **4j**.

Temperatura reakcije: 115 °C.

Iskorištenje: 0,245 g (65 %).

Ulje.

IR (KBr): ν_{max} 3296, 3064, 2980, 1735, 1633, 1564, 1449, 1374, 1284, 1172, 1077, 698, 643 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 8,47 (t, 1H, 1'(amid), J = 4,34 Hz), 7,75–7,49 (m, 18H, arom.), 7,37–7,22 (m, 5H, arom.), 6,84 (d, 1H, 3, J = 8,31 Hz), 6,36 (t, 1H, 1'(urea), J = 5,54 Hz), 5,32 (d, 1H, 2, J = 8,01 Hz), 4,10–3,78 (m, 6H, 3', 2"), 3,32–3,21 (m, 4H, 2'), 1,43, 1,37 (2d, 6H, 3", J = 7,32 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 196,04 (10"), 173,93, 173,85 (1"), 171,40 (1), 157,44 (4), 141,47, 141,39, 140,72, 140,70, 140,66, 137,62, 137,39 (5, 4", 8", 11",), 133,20, 132,31, 132,27, 129,28, 128,94, (5"–7", 9", 14"), 130,08, 129,04 (12", 13", 15", 16"), 127,64, 127,01 (6, 7, 9, 10), 127,68 (8), 64,59, 63,32 (3'), 56,97 (2), 44,75, 44,68 (2"), 38,59, 38,03 (2'), 18,99, 18,89 (3").

13-(3-Benzoilfenil)-5-benzil-4,7,12-triokso-11-oksa-3,6,8-triazatetradecil 2-(3-benzoil-fenil)propanoat (**12d**)

Količina reaktanta: 0,148 g spoja **4m**.

Temperatura reakcije: 115 °C.

Iskorištenje: 0,303 g (79 %).

Ulje.

IR (KBr): ν_{max} 3302, 3063, 2979, 2938, 1732, 1663, 1646, 1569, 1554, 1451, 1380, 1284, 1205, 1177, 1078, 956, 823, 722, 644 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 8,14 (t, 1H, 1'(amid), J = 5,25 Hz), 7,74–7,48 (m, 18H, arom.), 7,22–7,09 (m, 5H, arom.), 6,22–6,18 (d+t, 2H, 3, 1' (urea)), 4,36 (q, 1H, 2, J = 6,44 Hz), 4,07–3,88 (m, 6H, 3', 2"), 3,31–3,14 (m, 4H, 5, 2'), 2,88–2,66 (m, 2H, 2'), 1,44–1,41 (m, 6H, 3").

^{13}C NMR (DMSO-d_6, δ ppm): 196,04, 196,01 (10"), 173,93 (1"), 172,59 (1), 157,73 (4), 141,48, 141,46, 138,17, 138,14, 137,62, 137,61, 137,39 (6, 4", 8", 11"), 133,20, 132,30, 129,29, 128,96 (5"–7", 9", 14"), 130,07, 129,03 (12", 13", 15", 16"), 129,67, 128,39 (7, 8, 10, 11), 126,59 (9), 64,54 63,54, 63,40 (3'), 54,69 (2), 44,74 (2"), 39,34 (5), 38,55, 37,90 (2'), 19,00 (3").

2.1.14. Sinteza 1-acil-4-cikloalkil, 1-acil-4-aril i 1-acil-4-hidroksisemikarbazida NSAID 13a-y

Metoda A. Odgovarajući 1-karbamoilbenzotriazol (**7a-f**, 1 mmol), hidrazid NSAID-a **11a-c** (1 mmol) i TEA (0,418 mL, 3 mmol) rastaljeni su na temperaturi 75-120 °C. Reakcijska smjesa miješana je na toj temperaturi 0,25 h, ohlađena i otopljena u etil-acetatu (20 mL). Organski sloj ekstrahiran je zaluženom vodom (3×20 mL, 10 kapi 5 % NaOH) i vodom (1×20 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom.

Metoda B. Odgovarajući 1-karbamoilbenzotriazol **7a-f** (1 mmol), hidrazid NSAID-a **11a-c** (1 mmol) i TEA (0,418 mL, 3 mmol) rastaljeni su na temperaturi 75–120 °C. Reakcijska smjesa miješana je na toj temperaturi 0,25 h te ohlađena. Nastali talog rastrljan je u smjesi acetona i zakiseljene vode (pH = 1) i odsisan.

Sirovi produkti **13a-s** pročišćeni su prekristalizacijom iz etera/petroletera.

Metoda C. 1-(*N*-Benziloksikarbamoil)benzotriazol **7g** (0,268 g, 1 mmol) i odgovarajući hidrazid NSAID-a **11a-c** (1 mmol) otopljeni su u dioksanu i miješani 6-8 h na 55 °C. Reakcijska smjesa je uparena pod sniženim tlakom i otopljena u etil-acetatu. Organski sloj ekstrahiran je zaluženom vodom (3×20 mL, 6 kapi 5 % NaOH) i vodom (1×20 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Sirovi produkti **13t-v** su prekristalizirani iz acetona/ petroletera (**13t**), etera/petroletera (**13v**) ili pročišćeni kromatografijom na koloni (**13u**).

Metoda D. Suspenzija *O*-benzilnog derivata (**13t-v**, 0,5 mmol) i 50 mg 10 %-tnog Pd/C u metanolu (10 mL) hidrogenirana je pod atmosferskim tlakom vodika na sobnoj temperaturi tijekom 1,5 h. Katalizator je odfiltriran, a otapalo upareno pod sniženim tlakom. Sirovi produkti su pročišćeni prekristalizacijom iz etera/petroletera (**13w**) ili rastrljavanjem u eteru (**13x**, **13y**).

4-Ciklopentil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13a)
Metoda A. Količine reaktanata: 0,230 g spoja **7a**, 0,220 g spoja **11a**.
Temperatura reakcije: 75 °C.

Iskorištenje: 0,212 g (64 %).

t_t 123–128 °C.

IR (KBr): ν_{max} 3355, 3254, 2956, 2871, 1684, 1650, 1557, 1513, 1465, 1315, 1243, 1151, 1079, 1008, 937, 850, 786, 655 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,63, 7,59, (2s, 2H, 1, 2), 7,15 (dd, 4H, arom, J = 7,55 Hz, 87,09 Hz), 6,50 (bs, 1H, 2'), 3,84–3,80 (m, 1H, 3'), 3,59 (q, 1H, 4, J = 7,02 Hz), 2,40 (d, 2H, 12, J = 7,17 Hz), 1,84–1,20 (m, 9H, 13, 4'–7'), 1,32 (d, 3H, 5, J = 7,02 Hz), 0,85 (d, 6H, 14,15, J = 6,58 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 175,27 (3), 159,64 (1'), 141,54, 141,08 (6, 9), 130,96, 129,18 (7, 8, 10, 11), 53,09 (3'), 46,42 (12), 44,76 (4), 34,86, 25,33 (4'–7'), 31,76 (13), 24,34 (14, 15), 20,37 (5).

4-Cikloheksil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13b)

Metoda A. Količine reaktanata: 0,244 g spoja **7b**, 0,220 g spoja **11a**.

Temperatura reakcije: 90 °C.

Iskorištenje: 0,276 g (80 %).

t_t 165–166 °C.

IR (KBr): ν_{max} 3307, 3253, 3019, 2986, 2951, 2930, 2852, 1708, 1683, 1641, 1562, 1531, 1506, 1453, 1322, 1253, 1226, 1153, 1079, 1056, 1023, 999, 936, 892, 850, 709, 634, 614 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,67, 7,64, (2d, 2H, 1, 2, J = 1,77 Hz), 7,15 (dd, 4H, arom., J = 7,76 Hz, 39,23 Hz), 5,71 (d, 1H, 2', J = 6,43 Hz), 3,59 (q, 1H, 4, J = 6,87 Hz), 3,39–3,28 (m, 2H, 3'), 2,40 (d, 2H, 12, J = 7,10 Hz), 1,84–1,75 (m, 1H, 13), 1,69–0,95 (m, 10H, 4'–8'), 1,32 (d, 3H, 5, J = 6,87 Hz), 0,85 (d, 6H, 14, 15, J = 6,65 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,56 (3), 157,60 (1'), 139,84, 139,36 (6, 9), 129,27, 127,47 (7, 8, 10, 11), 48,28 (3'), 44,70 (12), 43,03 (4), 33,42, 33,38, 24,90 (4', 5', 7', 8'), 30,09 (13), 25,65 (6'), 22,64 (14, 15), 18,64 (5).

4-Cikloheksilmetil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13c)

Metoda B. Količine reaktanata: 0,258 g spoja **7c**, spoja **11a**.

Temperatura reakcije: 75 °C.

Iskorištenje: 0,338 g (94 %).

t_t 172–174 °C.

IR (KBr): ν_{max} 3339, 3236, 3122, 2952, 2926, 2849, 1699, 1615, 1565, 1476, 1448, 1368, 1263, 1247, 1175, 1084, 1019, 854, 780, 651, 618 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,65 7,66 (2s, 2H, 1, 2), 7,15 (dd, 4H, arom., J = 7,74 Hz, 40,61 Hz), 5,99 (t, 1H, 2', J = 4,78 Hz), 3,59 (q, 1H, 4, J = 6,86 Hz), 2,82 (t, 2H, 3', J = 6,22 Hz), 2,40 (d, 2H, 12, J = 7,02 Hz), 1,84–1,75 (m, 1H, 13), 1,66–0,78 (m, 11H, 4'–9'), 1,33 (d, 3H, 5, J = 6,53 Hz), 0,85 (d, 6H, 14, 15, J = 5,87 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,66 (3), 158,47 (1'), 139,79, 139,35 (6, 9), 129,22, 127,50 (7, 8, 10, 11), 45,80 (3'), 44,71 (12), 43,06 (4), 38,37 (4'), 30,67, 30,65, 25,88 (5', 6', 8', 9'), 30,09 (13), 26,56 (7'), 22,64 (14, 15), 18,69 (5).

4-Benzil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13d)

Metoda B. Količine reaktanata: 0,252 g spoja **7d**, 0,220 g spoja **11a**.

Temperatura reakcije: 110 °C.

Iskorištenje: 0,293 g (83 %).

t_t 173–175 °C.

IR (KBr): ν_{max} 3316, 3222, 3123, 3028, 2954, 1654, 1611, 1566, 1469, 1454, 1364, 1236, 1171, 1077, 851, 749, 699, 668 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,70, 7,85 (2s, 2H, 1, 2), 7,32–7,05 (m, 9H, arom.), 6,70 (t, 1H, 2', J = 6,01 Hz), 4,22 (d, 2H, 3', J = 6,01 Hz), 3,59 (q, 1H, 4, J = 6,94 Hz), 2,39 (d, 2H, 12, J = 6,94 Hz), 1,83–1,74 (m, 1H, 13), 1,34 (d, 3H, 5, J = 6,94 Hz), 0,84 (d, 6H, 14, 15, J = 6,48 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,83 (3), 158,59 (1'), 140,90 (4'), 139,78, 139,34 (6, 9), 129,21, 128,58, 127,55, 127,35, (7, 8, 10, 11, 5', 6', 8', 9'), 127,00 (7'), 44,70 (12), 43,11 (4), 43,07 (3'), 30,08 (13), 22,64 (14, 15), 18,81 (5).

4-Feniletil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13e)

Metoda A. Količine reaktanata: 0,252 g spoja **7e**, 0,220 g spoja **11a**.

Temperatura reakcije: 115 °C.

Iskorištenje: 0,268 g (73 %).

t_t 173–175 °C.

IR (KBr): ν_{max} 3566, 3434, 3312, 3026, 2953, 2926, 2869, 1648, 1587, 1557, 1514, 1497, 1455, 1374, 1253, 1168, 1074, 1054, 999, 934, 853, 796, 746, 700, 547 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,66, 7,79 (2s, 2H, 1, 2), 7,38–7,00 (m, 9H, arom.), 6,15 (t, 1H, 2', J = 5,52 Hz), 3,59 (q, 1H, 4, J = 6,81 Hz), 3,22 (q, 2H, 3', J = 6,49 Hz), 2,67 (t, 2H, 4', J = 7,14 Hz), 2,40 (d, 2H, 12, J = 6,81 Hz), 1,85–1,76 (m, 1H, 13), 1,34 (d, 3H, 5, J = 7,14 Hz), 0,86 (d, 6H, 14, 15, J = 6,49 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,72 (3), 158,36 (1'), 139,99, 139,79, 139,33 (6, 9, 5'), 129,21, 129,10, 128,78, 127,54, (7, 8, 10, 11, 6', 7', 9', 10'), 126,48 (8'), 44,70 (12), 43,06 (4), 41,32 (3'), 36,38 (4'), 30,08 (13), 22,64 (14, 15), 18,78 (5).

4-Benzhidril-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13f)

Metoda A. Količine reaktanata: 0,328 g spoja **7f**, 0,220 g spoja **11a**.

Temperatura reakcije: 120 °C.

Iskorištenje: 0,314 g (73 %).

t_t 151–154 °C.

IR (KBr): ν_{max} 3343, 3202, 3028, 2953, 2869, 1647, 1616, 1538, 1495, 1456, 1366, 1268, 1230, 1168, 1077, 1052, 1028, 1004, 942, 848, 746, 700, 669, 631 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,76, 7,84 (2s, 2H, 1, 2), 7,34–7,05 (m, 14H, arom.), 6,95 (bs, 1H, 2'), 5,91 (d, 1H, 3', J = 8,38 Hz), 3,58 (q, 1H, 4, J = 6,85 Hz), 2,39 (d, 2H, 12, J = 7,11 Hz), 1,83–1,74 (m, 1H, 13), 1,33 (d, 3H, 5, J = 6,85 Hz), 0,84 (d, 6H, 14, 15, J = 6,60 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,70 (3), 157,58 (1'), 143,51, 143,46 (4', 10'), 139,83, 139,26 (6, 9), 129,25, 128,81, 128,79, 127,49, 127,47, 127,44 (7, 8, 10, 11, 5', 6', 8', 9', 11', 12', 14', 15'), 127,35, 127,31 (7', 13'), 57,14 (3'), 44,71 (12), 43,05 (4), 30,06 (13), 22,65 (14, 15), 18,78 (5).

4-Ciklopentil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13g)

Metoda A. Količine reaktanata: 0,230 g spoja **7a**, 0,256 g spoja **11b**.

Temperatura reakcije: 90 °C.

Iskorištenje: 0,293 g (80 %).

t_t 133–135 °C.

IR (KBr): ν_{max} 3414, 3281, 3237, 3101, 3025, 2961, 2910, 2869, 1696, 1627, 1638, 1582, 1556, 1532, 1489, 1455, 1443, 1311, 1244, 1213, 1162, 1146,1131, 1073, 942, 913, 880, 774, 760, 691, 618 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,69, 7,63 (2s, 2H, 1, 2), 7,42–6,82 (m, 9H, arom.), 5,94 (d, 1H, 2', J = 6,73 Hz), 3,89–3,78 (m, 2H, 3'), 3,63 (q, 1H, 4, J = 6,73 Hz), 1,79–1,22 (m, 8H, 4'–7'), 1,32 (d, 3H, 5, J = 6,73 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,08 (3), 157,89 (1'), 156,99, 156,94, 144,29 (6, 10, 12), 130,49, 119,01 (13, 14, 16, 17), 130,23, 123,87 , 122,93, 118,17, 117,12 (7–9, 11, 15), 51,40 (3'), 43,25 (4), 33,13, 24,91 (4'–7'), 18,66 (5).

4-Cikloheksil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13h)

Metoda A. Količine reaktanata: 0,244 g spoja **7b**, 0,256 g spoja **11b**.

Temperatura reakcije: 90 °C.

Iskorištenje: 0,229 g (60 %).

t_t 144–146 °C.

IR (KBr): ν_{max} 3343, 3215, 2933, 2853, 1695, 1618, 1580, 1555, 1490, 1472, 1448, 1314, 1273, 1235, 1213, 1166, 1074, 1062, 1025, 944, 887, 748, 689, 640 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,70, 7,66 (2s, 2H, 1, 2), 7,42–6,83 (m, 9H, arom.), 5,84 (d, 1H, 2', J = 7,00 Hz), 3,63 (q, 1H, 4, J = 6,73 Hz), 3,39–3,33 (m, 2H, 3'), 1,71–1,00 (m, 10H, 4'–8'), 1,33 (d, 3H, 5, J = 7,00 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,08 (3), 157,55 (1'), 156,99, 156,94, 144,29 (6, 10, 12), 130,49, 119,01 (13, 14, 15, 16), 130,24, 123,87 , 122,95, 118,16, 117,12 (7–9, 11, 15), 48,33 (3'), 43,24 (4), 33,41, 24,91 (4', 5', 7', 8'), 25,65 (6'), 18,66 (5).

4-Cikloheksanmetil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13i)

Metoda A. Količine reaktanata: 0,258 g spoja **7c**, 0,256 g spoja **11b**.

Temperatura reakcije: 75 °C.

Iskorištenje: 0,198 g (50 %).

t_t 104–105 °C.

IR (KBr): ν_{max} 3339, 3230, 3115, 3026, 2924, 2850, 1701, 1619, 1582, 1568, 1490, 1447, 1373, 1246, 1212, 1166, 1146, 1074, 1024, 1012, 962, 933, 888, 749, 691, 668 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,70, 7,70 (2s, 2H, 1, 2), 7,43–6,84 (m, 9H, arom.), 6,09 (t, 1H, 2', J = 5,20 Hz), 3,63 (q, 1H, 4, J = 6,82 Hz), 2,84 (t, 2H, 3', J = 6,17 Hz), 1,62–0,79 (m, 11H, 4'–9'), 1,34 (d, 3H, 5, J = 6,82 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,18 (3), 158,43 (1'), 157,03, 156,91, 144,29 (6, 10, 12), 130,19, 118,98 (13, 14, 16, 17), 130,21, 123,85 , 122,98, 118,23, 117,14 (7–9, 11, 15), 45,82 (3'), 43,29 (4), 38,38 (4'), 30,68, 25,88 (4', 5', 8', 9'), 26,56 (7'), 18,70 (5).

4-Benzil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13j)

Metoda A. Količine reaktanata: 0,252 g spoja **7d**, 0,256 g spoja **11b**.

Temperatura reakcije: 110 °C.

Iskorištenje: 0,233 g (60 %).

t_t 154–156 °C.

IR (KBr): ν_{max} 3323, 3266, 3209, 3109, 3028, 1654,1621, 1584, 1565, 1490, 1453, 1369, 1315, 1278, 1249, 1235, 1162, 1143, 1071, 935,876, 751, 694, 644 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,74, 7,89 (2s, 2H, 1, 2), 7,41–6,82 (m, 14H, arom.), 6,76 (t, 1H, 2', J = 5,58 Hz), 4,22 (d, 2H, 3', J = 5,58 Hz), 3,63 (q, 1H, 4, J = 6,50 Hz), 1,34 (d, 3H, 5, J = 6,81 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,35 (3), 158,54 (1'), 157,03, 156,90, 144,28 (6, 10, 12), 140,91 (4'), 130,49, 128,59, 127,34, 119,00 (13, 14, 16, 17, 5', 6', 8', 9'), 130,20, 127,00, 123,84, 123,02, 118,29, 117,14 (7–9, 11, 15, 7'), 43,36 (4), 43,08 (3'), 18,77 (5).

4-Feniletil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13k)

Metoda B. Količine reaktanata: 0,252 g spoja **7e**, 0,256 g spoja **11b**.

Temperatura reakcije: 115 °C.

Iskorištenje: 0,254 g (63 %).

t_t 116–118 °C.

IR (KBr): ν_{max} 3367, 3236, 3026, 2942, 1682, 1642, 1583, 1560, 1542, 1488, 1454, 1379, 1310, 1247, 1211, 1162, 1075, 1012, 944, 916, 866, 796, 747, 698, 637 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,70, 7,82 (2s, 2H, 1, 2), 7,42–6,84 (m, 14H, arom.), 6,21 (t, 1H, 2', J = 5,36 Hz), 3,63 (q, 1H, 4, J = 6,80 Hz), 3,23 (q, 2H, 3', J = 6,59 Hz), 2,68 (t, 2H, 4', J = 7,21 Hz), 1,34 (d, 3H, 5, J = 7,00 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,23 (3), 158,31 (1'), 157,03, 156,90, 144,29 (6, 10, 12), 139,99 (5'), 130,49, 129,11, 128,79, 119,00, (13, 14, 16, 17, 6', 7', 9', 10'), 130,21, 126,48, 123,85, 123,02, 118,26, 117,14 (7–9, 11, 15, 8'), 43,29 (4), 41,32 (3'), 36,38 (4'), 18,78 (5).

4-Benzhidril-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13l)

Metoda A. Količine reaktanata: 0,328 g spoja **7f**, 0,256 g spoja **11b**.

Temperatura reakcije: 120 °C.

Iskorištenje: 0,219 g (47 %).

t_t 126–131 °C.

IR (KBr): ν_{max} 3350, 3236, 3031, 2973, 1655, 1613, 1587, 1560, 1488, 1448, 1247, 1210, 1164, 1070, 1052, 1030, 949, 912, 753, 698, 668, 634, 614 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,78, 7,85 (2s, 2H, 1, 2), 7,38–6,98 (m, 19H, arom.), 6,83 (d, 1H, 2', J = 7,36 Hz), 5,91 (d, 1H, 3', J = 8,83 Hz), 3,64 (q, 1H, 4, J = 6,31 Hz), 1,33 (d, 3H, 5, J = 6,75 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,70 (3), 157,02 (1'), 156,50 156,47, 143,70 (6, 10, 12), 142,96, 143,03 (4', 10'), 129,97, 118,53 (13, 14, 16, 17), 129,73, 123,35, 122,43, 117,71, 116,65 (7–9, 11, 15), 128,37, 128,33, 126,97, 126,93 (5', 6', 8', 9', 11', 12', 14', 15'), 126,85, 126,81 (7',13'), 56,71 (3'), 42,80 (4), 18,24 (5).

4-Ciklopentil-1-[2-(3-benzilfenil)propanoil]semikarbazid (13m)

Metoda A. Količine reaktanata: 0,230 g spoja **7a**, 0,254 g spoja **11c**.

Temperatura reakcije: 75 °C.

Iskorištenje: 0,259 g (71 %).

t_t 133–134 °C.

IR (KBr): ν_{max} 3279, 3027, 2962, 2872, 1710, 1646, 1555, 1494, 1452, 1372, 1242, 1191, 1152, 1077, 1010, 941, 728, 698, 638cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,67, 7,60 (2s, 2H, 1, 2), 7,31–7,05 (m, 9H, arom.), 5,90 (d, 1H, 2', J = 6,27 Hz), 3,91 (s, 2H, 12), 3,82 (m, 1H, 3'), 3,59 (q, 1H, 4, J = 6,86 Hz), 1,76–1,22 (m, 8H, 4'–7'), 1,32 (d, 3H, 5, J = 7,05 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,40 (3), 157,93 (1'), 142,22, 141,61, 141,55 (6, 10, 13), 129,14, 128,86 (14, 15, 17, 18), 128,76, 128,25, 127,48, 126,42, 125,45 (7–9, 11, 16), 51,38 (3'), 43,36 (4), 41,65 (12), 33,13, 23,62 (4'–7'), 18,73 (5).

4-Cikloheksil-2-[2-(3-benzilfenil)propanoil]semikarbazid (13n)

Metoda A. Količine reaktanata: 0,244 g spoja **7b**, 0,254 g spoja **11c**.

Temperatura reakcije: 90 °C.

Iskorištenje: 0,334 g (85 %).

t_t 143–144 °C.

IR (KBr): ν_{max} 3327, 3216, 3027, 2932, 2853, 1658, 1617, 1589, 1477, 1452, 1255, 1232, 1182, 1074, 1065, 943, 891, 769, 751, 701, 639,618 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,68, 7,64 (2s, 2H, 1, 2), 7,30–7,05 (m, 9H, arom.), 5,80 (d, 1H, 2', J = 7,29 Hz), 3,91 (s, 2H, 12), 3,59 (q, 1H, 4, J = 6,97 Hz), 3,38–3,29 (m, 1H, 3'), 1,70–0,97 (m, 10H, 4'–8'), 1,32 (d, 3H, 5, J = 6,97 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,40 (3), 157,60 (1'), 142,22, 141,61, 141,55 (6, 10, 13), 129,14, 128,86 (14, 15, 17, 18), 128,77, 128,24, 127,48, 126,42, 125,45 (7–9, 11, 16), 48,31 (3'), 43,36 (4), 41,65 (12), 33,38, 24,89 (4', 5', 7', 8'), 25,66 (6'), 18,73 (5).

4-Cikloheksilmetil-1-[2-(3-benzilfenil)propanoil]semikarbazid (13o)

Metoda B. Količine reaktanata: 0,258 g spoja **7c**, 0,254 g spoja **11c**.

Temperatura reakcije: 75 °C.

Iskorištenje: 0,268 g (68 %).

t_t 134–137 °C.

IR (KBr): ν_{max} 3344, 3216, 3027, 2919, 2850, 1663, 1622, 1589, 1474, 1450, 1370, 1273, 1257, 1239, 1184, 1075, 1030, 963, 746, 723, 702, 652, 618 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,67, 7,66 (2s, 2H, 1, 2), 7,31–7,05 (m, 9H, arom.), 6,04 (t, 1H, 2', J = 5,23 Hz), 3,91 (s, 2H, 12), 3,59 (q, 1H, 4, J = 6,82 Hz), 2,82 (t, 2H, 3', J = 6,14 Hz), 1,66–0,78 (m, 11H, 4'–9'), 1,32 (d, 3H, 5, J = 6,82 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,48 (3), 158,47 (1'), 142,21, 141,62, 141,52 (6, 10, 13), 129,13, 128,86 (14, 15, 17, 18), 128,74, 128,24, 127,46, 126,41, 125,47 (7–9, 11, 16), 45,81 (3'), 43,38 (4), 41,65 (12), 38,36 (4'), 30,66, 25,88 (5', 6', 8', 9'), 26,55 (7'), 18,76 (5).

4-Benzil-1-[2-(3-benzilfenil)propanoil]semikarbazid (13p)

Metoda B. Količine reaktanata: 0,252 g spoja **7d**, 0,256 g spoja **11c**.

Temperatura reakcije: 110 °C.

Iskorištenje: 0,325 g (84 %).

t_t 150–154 °C.

IR (KBr): ν_{max} 3331, 3261, 3211, 3028, 2986, 1707, 1660, 1620, 1563, 1494, 1453, 1431, 1371, 1246, 1174, 1075, 750, 723, 698, 642, 618 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,72, 7,86 (2s, 2H, 1, 2), 7,32–7,04 (m, 14H, arom.), 6,73 (t, 1H, 2', J = 6,19 Hz), 4,22 (d, 2H, 3', J = 5,77 Hz), 3,89 (s, 2H, 12), 3,60 (q, 1H, 4, J = 7,01 Hz), 1,34 (d, 3H, 5, J = 7,01 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,67 (3), 158,59 (1'), 142,23, 141,63, 141,52 (6, 10, 13), 140,92 (4'), 129,15, 128,87, 128,59, 127,35 (14, 15, 17, 18, 5', 6', 8', 9'), 128,74, 128,28, 127,46, 127,01, 126,41, 125,55 (7–9, 11, 16, 7'), 43,46 (4), 43,08 (3'), 41,64 (12), 18,88 (5).

4-Feniletil-1-[2-(3-benzilfenil)propanoil]semikarbazid (13r)

Metoda A. Količine reaktanata: 0,252 g spoja **7e**, 0,254 g spoja **11c**.

Temperatura reakcije: 115 °C.

Iskorištenje: 0,303 g (73 %).

t_t 118–120 °C.

IR (KBr): ν_{max} 3365, 3301, 3239, 3027, 2939, 1686, 1649, 1601, 1555, 1495, 1455, 1379, 1269, 1242, 1154, 1073, 945, 751, 726, 699, 661 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,66, 7,78 (2s, 2H, 1, 2), 7,29–7,04 (m, 14H, arom.), 6,17 (t, 1H, 2', J = 5,61 Hz), 3,90 (s, 2H, 12), 3,58 (q, 1H, 4, J = 6,83 Hz), 3,21 (q, 2H, 3', J = 6,58 Hz), 2,65 (t, 2H, 4', J = 7,31 Hz), 1,32 (d, 3H, 5, J = 7,07 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,55 (3), 158,37 (1'), 142,23, 141,63, 141,52 (6, 10, 13), 140,00 (5'), 129,15, 129,11, 128,87, 128,79 (14, 15, 17, 18, 6', 7', 9', 10'), 128,75, 128,28, 127,46, 126,48, 126,41, 125,54 (7–9, 11, 16, 8'), 43,41 (4), 41,65 (12), 41,33 (3'), 36,39 (4'), 18,87 (5).

4-Benzhidril-1-[2-(3-benzilfenil)propanoil]semikarbazid (13s)

Metoda A. Količine reaktanata: 0,328 g spoja **7f**, 0,254 g spoja **11c**.

Temperatura reakcije: 120 °C.

Iskorištenje: 0,339 g (71 %).

t_t 166–170 °C.

IR (KBr): ν_{max} 3346, 3231, 3028, 1981, 1662, 1611, 1590, 1560, 1494, 1472, 1453, 1372, 1258, 1236, 1078, 1030, 948, 751, 700, 668, 634, 613 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,78, 7,84 (2s, 2H, 1, 2), 7,33–7,00 (m, 19H, arom.), 6,99 (d, 1H, 2', J = 8,85 Hz), 5,91 (d, 1H, 3', J =8,38 Hz), 3,89 (s, 2H, 12), 3,60 (q, 1H, 4, J = 6,99 Hz), 1,33 (d, 3H, 5, J = 6,99 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,67 (3), 157,56 (1'), 143,52, 143,47 (4', 10'), 142,13, 141,59, 141,56, (6, 10, 13), 129,14, 128,86, 128,82, 128,80, 127,45 (14, 15, 17, 18, 5', 6', 8', 9', 11', 12', 14', 15'), 128,22, 127,34, 127,32, 126,40, 125,47 (7–9, 11, 16, 7', 13'), 57,16 (3'), 43,48 (4), 41,63 (12), 18,84 (5).

4-Benziloksi-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13t)

Metoda C. Količina reaktanta: 0,220 g spoja **11a**.

Trajanje reakcije: 6 h.

Iskorištenje: 0,336 g (91 %).

t_t 87 °C (raspad).

IR (KBr): ν_{max} 3472, 3319, 3240, 3114, 3064, 3031, 2953, 2867, 1714, 1698, 1681, 1638, 1594, 1548, 1514, 1455, 1366, 1349, 1226, 1147, 1082, 994, 933, 910, 850, 802, 751, 698, 620, 592, 545 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,73, 9,47, 8,71 (3s, 3H, 1, 2, 2'), 7,43–7,32 (m, 5H, arom.), 7,17 (dd, 4H, arom., J = 8,03 Hz, 45,43 Hz), 4,74 (s, 2H, 3'), 3,62 (q, 1H, 4, J = 6,81 Hz), 2,40 (d, 2H, 12, J = 6,99 Hz), 1,87–1,76 (m, 1H, 13), 1,34 (d, 3H, 5, J = 7,16 Hz), 0,85 (d, 6H, 14, 15, J = 6,46 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,55 (3), 159,52 (1'), 139,77, 139,25 (6, 9), 136,90 (4'), 129,17, 129,12, 128,61, 127,62 (7, 8, 10, 11, 5', 6', 8', 9'), 128,42 (7'), 77,90 (3'), 44,71 (12), 43,05 (4), 30,10 (13), 22,65 (14,15), 18,93 (5).

4-Benziloksi-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13u)

Metoda C. Količina reaktanta: 0,256 g spoja **11b**.

Trajanje reakcije: 7 h.

Iskorištenje: 0,369 g (91 %).

Ulje.

IR (KBr): ν_{max} 3237, 3063, 3034, 2979, 2934, 2877, 1668, 1583, 1488, 1455, 1371, 1312, 1244, 1211, 1163, 1073, 942, 914, 753, 695, 614 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,74, 9,47, 8,70 (3s, 3H, 1, 2, 2'), 7,41–6,83 (m, arom., 14H), 4,75 (s, 2H, 3'), 3,66 (q, 1H, 4, J = 6,94 Hz), 1,35 (d, 3H, 5, J = 6,94 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,59 (3), 158,95 (1'), 156,57, 156,38, 143,70 (6, 10, 12), 136,37 (4'), 129,98, 128,62, 128,11, 118,48 (13, 14, 16, 17, 5', 6', 8', 9'), 129,65, 127,93, 123,32, 122,59, 117,87, 116,65 (7–9, 11, 15, 7'), 77,42 (3'), 42,80 (4), 18,37 (5).

4-Benziloksi-1-[2-(3-benzilfenil)propanoil]semikarbazid (13v)

Metoda C. Količina reaktanta: 0,254 g spoja **11c**.

Trajanje reakcije: 8 h.

Iskorištenje: 0,201 g (50 %).

t_t 104–105 °C.

IR (KBr): ν_{max} 3306, 3235, 3029, 2925, 2880, 1691, 1654, 1599, 1520, 1493, 1454, 1382, 1366, 1356, 1273, 1238, 1152, 1074, 1032, 1007, 947, 788, 751, 727, 699, 668, 555, 532, 497 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,72, 9,46, 8,69 (3s, 3H, 1, 2, 2'), 7,37–7,02 (m, arom., 14H), 4,72 (s, 2H, 3'), 3,88 (s, 2H, 12), 3,59 (q, 1H, 4, J = 6,46 Hz), 1,31 (d, 3H, 5, J = 6,85 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,40 (3), 159,52 (1'), 142,13, 141,65, 141,47 (6, 10, 13), 136,90 (4'), 129,16, 129,13, 128,87, 128,61 (14, 15, 17, 18, 5', 6', 8', 9'), 128,70, 128,43, 128,34, 127,44, 126,41, 125,61 (7–9, 11, 16, 7'), 77,90 (3'), 43,39 (4), 41,65 (12), 19,00 (5).

4-Hidroksi-1-[2-(4-izobutilfenil)propanoil]semikarbazid (13w)

Metoda D. Količina reaktanta: 0,185 g spoja **13t**.

Iskorištenje: 0,134 g (96 %).

t_t 123–125 °C.

IR (KBr): ν_{max} 3330, 3235, 3027, 2954, 2924, 2869, 1704, 1652, 1556, 1514, 1465, 1383, 1320, 1262, 1141, 1078, 1002, 935, 850, 766, 727, 653, 531 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,65, 8,70, 8,64, 8,41 (4s, 4H, 1, 2, 2', 3'), 7,15 (dd, 4H, arom, J = 7,92 Hz, 46,47 Hz), 3,61 (q, 1H, 4, J = 6,89 Hz), 2,40 (d, 2H, 12, J = 7,11 Hz), 1,85–1,76 (m, 1H, 13), 1,33 (d, 3H, 5, J = 6,89 Hz), 0,85 (d, 6H, 14, 15, J = 6,45 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,45 (3), 160,82 (1'), 139,71, 139,33 (6, 9), 129,14, 127,62 (7, 8, 10,11), 44,71 (12), 43,00 (4), 30,09 (13), 22,65 (14,15), 19,01 (5).

4-Hidroksi-1-[2-(3-fenoksifenil)propanoil]semikarbazid (13x)

Metoda D. Količina reaktanta: 0,203 g spoja **13u**.

Iskorištenje: 0,118 g (75 %).

t_t 150 °C (raspad).

IR (KBr): ν_{max} 3298, 3214, 3026, 2983, 2941, 1685, 1643, 1582, 1567, 1489, 1446, 1362, 1313, 1268, 1232, 1212, 1161, 1140, 1080, 1023, 969, 939, 876, 788, 756, 693, 649, 596, 558, 523 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,68, 8,72, 8,65, 8,45 (4s, 4H, 1, 2, 2', 3'), 7,42–6,82 (m, arom., 9H), 3,65 (q, 1H, 4, J = 6,95 Hz), 1,33 (d, 3H, 5, J = 6,95 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,98 (3), 160,78 (1'), 157,06, 156,84, 144,29 (6, 10, 12), 130,49, 118,98 (13, 14,16, 17), 130,13, 123,81, 123,10, 118,36, 117,11 (7–9, 11, 15), 43,24 (4), 18,95 (5).

4-Hidroksi-1-(2-(3-benzilfenil)propanoil]semikarbazid (13y)

Metoda D. Količina reaktanta: 0,202 g spoja **13v**.

Iskorištenje: 0,150 g (96 %).

t_t 123–125 °C.

IR (KBr): ν_{max} 3288, 3060, 3028, 2979, 2938, 1687, 1644, 1562, 1516, 1494, 1483, 1451, 1380, 1263, 1159, 1144, 1100, 1077, 1061, 1027, 938, 782, 721, 700, 596, 533 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,66, 8,73, 8,66, 8,43 (4s, 4H, 1, 2, 2', 3'), 7,32–7,05 (m, arom., 9H), 3,92 (s, 2H, 12), 3,62 (q, 1H, 4, J = 6,81 Hz), 1,34 (d, 3H, 5, J = 7,01 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,29 (3), 160,82 (1'), 142,22, 141,66, 141,45 (6, 10, 13), 129,16, 128,87 (14, 15, 17, 18), 128,68, 128,34, 127,38, 126,40, 125,60 (7–9, 11, 16), 43,34 (4), 41,65 (12), 19,08 (5).

2.1.15. Sinteza 1-acil-5-benziloksi i 1-acil-5-hidroksikarbamoil karbazida NSAID 14a-f

Metoda A. Odgovarajući hidrazid NSAID-a **11a-c** (1 mmol) i 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazid (**9**) (0,326 g, 1 mmol) rastaljeni su na 93 °C. Reakcijska smjesa je na toj temperaturi miješana 0,3 h, ohlađena i otopljena u etil-acetatu. Organski sloj ekstrahiran je zaluženom vodom (3×20 mL, 6 kapi 5 % NaOH) i vodom (1×20 mL), osušen nad bezvodnim natrijevim sulfatom, profiltriran i uparen pod sniženim tlakom. Sirovi produkti **14a-c** pročišćeni su kromatografijom na koloni uz pokretnu fazu diklormetan/metanol 9,5:0,5 i prekristalizacijom iz etera/petroletera.

Metoda B. Suspenzija *O*-benzilnog derivata (**14a-c,** 0,5 mmol) i 50 mg 10 %-tnog Pd/C u metanolu (10 mL) hidrogenirana je pod atmosferskim tlakom vodika na sobnoj temperaturi

tijekom 2 h. Katalizator je odfiltriran, a otapalo upareno pod sniženim tlakom. Sirovi produkti su pročišćeni rastrljavanjem u eteru.

5-(Benziloksikarbamoil)-1-[2-(4-izobutilfenil)propanoil]karbazid (14a)

Metoda A. Količina reaktanta: 0,220 g spoja **11a**.

Iskorištenje: 0,304 g (71 %).

t_t 85–90 °C.

IR (KBr): ν_{max} 3266, 3032, 2955, 2930, 2869, 1677, 1513, 1466, 1455, 1366, 1309, 1271, 1212, 1188, 1075, 1030, 1002, 934, 909, 849, 751, 699, 620, 547 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,75, 9,43, 8,54, 8,15, 8,03 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–7,30 (m, 5H, arom., 8'–12'), 7,16 (dd, 4H, arom., J = 7,99 Hz, 46,08), 4,77 (s, 2H, 6'), 3,62 (q, 1H, 4, J = 6,94 Hz), 2,41 (d, 2H, 12, J = 7,15 Hz), 1,85–1,76 (m, 1H, 13), 1,35 (d, 3H, 5, J = 6,94 Hz), 0,86 (d, 6H, 14, 15, J = 6,52 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,52 (3), 160,03, 158,37 (1', 4'), 139,75, 139,34, (6, 9), 136,97 (7'), 129,17, 129,14, 128,61, 127,59 (7, 8, 10, 11, 8', 9', 11', 12'), 128,41 (10'), 77,87 (6'), 44,71 (12), 43,00 (4), 30,09 (13), 22,65 (14, 15), 18,85 (5).

5-Benziloksikarbamoil-1-[2-(3-fenoksifenil)propanoil]karbazid (14b)

Metoda A. Količina reaktanta: 0,256 g spoja **11b**.

Iskorištenje: 0,338 g (73 %).

t_t 72–76 °C.

IR (KBr): ν_{max} 3251, 3062, 3034, 2971, 2933, 2873, 1677, 1583, 1523, 1488, 1455, 1369, 1312, 1243, 1211, 1163, 1073, 1023, 1002, 942, 914, 816, 752, 694, 614 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,78, 9,43, 8,55, 8,18, 8,05 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–6,82 (m, 14H, arom.), 4,77 (s, 2H, 6'), 3,65 (q, 1H, 4, J = 6,97 Hz), 1,34 (d, 3H, 5, J = 7,12 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,03 (3), 160,03, 158,32 (1', 4'), 157,05, 156,87, 144,28 (6, 10, 12), 136,97 (7'), 130,50, 129,14, 128,61, 118,99 (13, 14, 16, 17, 8', 9', 11', 12'), 130,16, 128,41, 123,83, 123,06, 118,33,117,13 (7–9, 11, 15, 10'), 77,88 (6'), 43,25 (4), 18,79 (5).

5-Benziloksikarbamoil-1-[2-(3-benzilfenil)propanoil]karbazid (14c)

Metoda A. Količina reaktanta: 0,254 g spoja **11c**.

Iskorištenje: 0,337 g (73 %).

t_t 73–77 °C.

IR (KBr): ν_{max} 3248, 3060, 3028, 2880, 2836, 1675, 1602, 1519, 1494, 1453, 1369, 1310, 1270, 1210, 1075, 1030, 974, 941, 908, 751, 699, 620, 556 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,76, 9,43, 8,55, 8,16, 8,03 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–7,05 (m, 14H, arom.), 4,78 (s, 2H, 6'), 3,92 (s, 2H, 12), 3,62 (q, 1H, 4, J = 6,77 Hz), 1,34 (d, 3H, 5, J = 7,02 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,35 (3), 160,02, 158,36 (1', 4'), 142,21, 141,64, 141,48 (6, 10, 13), 136,96 (7'), 129,15, 128,87, 128,61 (14, 15, 17, 18, 8', 9', 11', 12'), 128,70, 128,41, 128,31, 127,42, 126,40, 125,58 (7–9, 11, 16, 10'), 77,88 (6'), 43,34 (4), 41,65 (12), 18,91 (5).

5-Hidroksikarbamoil-1-[2-(4-izobutilfenil)propanoil]karbazid (14d)

Metoda B. Količina reaktanta: 0,214 g spoja **14a**.

Iskorištenje: 0,120 g (71 %).

t_t 145–148 °C (raspad).

IR (KBr): ν_{max} 3261, 2955, 2928, 2870, 1670, 1514, 1466, 1383, 1367, 1281, 1206, 1077, 1002, 934, 850, 783 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,73, 8,66, 8,57, 8,23, 8,08, 7,92 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,15 (dd, 4H, arom., J = 7,93 Hz, 95,99 Hz), 3,61 (q, 1H, 4, J = 7,01 Hz), 2,40 (d, 2H, 12, J = 7,28 Hz), 1,82–1,78 (m, 1H, 13), 1,33 (d, 3H, 5, J = 7,01 Hz), 0,85 (d, 6H, 14, 15, J = 6,47 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,90 (3), 160,70, 157,83 (1', 4'), 139,24, 138,85 (6, 9), 128,6, 127,07 (7, 8, 10, 11), 44,22 (12), 42,49 (4), , 29,55 (13), 22,14 (14, 15), 18,35 (5).

5-Hidroksikarbamoil-1-[2-(3-fenoksifenil)propanoil]karbazid (14e)

Metoda B. Količina reaktanta: 0,232 g spoja **14b**.

Iskorištenje: 0,159 g (85 %).

t_t 100–103 °C (raspad).

IR (KBr): ν_{max} 3263, 2978, 2925, 2855, 1670, 1583, 1540, 1488, 1456, 1445, 1376, 1313, 1244, 1210, 1163, 1074, 943, 917, 755, 692 cm^{-1}.

^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,80, 8,67, 8,29, 8,14, 7,96 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,42–6,81 (m, 9H, arom.), 3,64 (q, 1H, 4, J = 7,03 Hz), 1,34 (d, 3H, 5, J = 6,95 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 172,92 (3), 161,24, 158,30 (1', 4'), 157,03, 156,88, 144,28, (6, 10, 12), 130,50, 119,01 (13, 14, 16, 17), 130,16, 123,84, 123,03, 118,30, 117,11 (7–9, 11, 15), 43,22 (4), 18,78 (5).

5-Hidroksikarbamoil-1-[2-(3-benzilfenil)propanoil]karbazid (14f)

Metoda B. Količina reaktanta: 0,231 g spoja **14c**.

Iskorištenje: 0,147 g (79 %).

t_t 101–105 °C.

IR (KBr): ν_{max} 3258, 3060, 3027, 2981, 2934, 1670, 1601, 1538, 1494,1453, 1375, 1321, 1195, 1075, 1030, 943, 784, 728, 699, 622, 557 cm^{-1}.

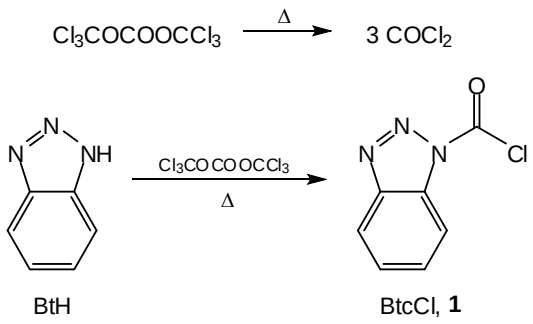
^{1}H NMR (DMSO-d_6, δ ppm, J/Hz): 9,78, 8,67, 8,28, 8,11, 7,95 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,31–7,04 (m, 9H, arom.), 3,91 (s, 2H, 12), 3,61 (q, 1H, 4, J = 6,88 Hz), 1,33 (d, 3H, 5, J = 7,14 Hz).

^{13}C NMR (DMSO-d_6, δ ppm): 173,23 (3), 161,24, 158,34 (1', 4'), 142,22, 141,64, 141,48 (6, 10, 13), 129,16, 128,88 (14, 15, 17, 18), 128,71, 128,29, 127,42, 126,41, 125,56 (7–9, 11, 16), 43,32 (4), 41,65 (12), 18,90 (5).

3. REZULTATI I RASPRAVA

3.1. SINTEZE

Početni spoj za pripravu derivata aminokiselina *N*-(1-benzotriazolkarbonil)-aminokiselina (*N*-Btc-aminokiselina) (**2**), 1-karbamoilbenzotriazola (**7**), 1-(1-benzotriazol-karbonil)-4-benziloksisemikarbazida (**9**) i NSAID benzotriazolida (**10**) bio je klorid 1-benzotriazolkarboksilne kiseline (BtcCl, **1**). Taj spoj dobiven je refluksiranjem benzotriazola i trifozgena u toluenskoj otopini (Shema 20) (Kalčić *et al*, 2003). Klorid **1** može se prirediti i iz benzotriazola i 20 %-tne otopine fozgena u toluenu ili uvođenjem plinovitog fozgena iz metalnog spremnika u toluensku otopinu benzotriazola (Butula *et al*, 1977). Zbog otežanog rukovanja i sigurnosti na radu u laboratoriju, danas se sve više upotrebljava čvrsti trifozgen (trimer fozgena) koji se zagrijavanjem razlaže u fozgen prema jednadžbi prikazanoj na Shemi 20.

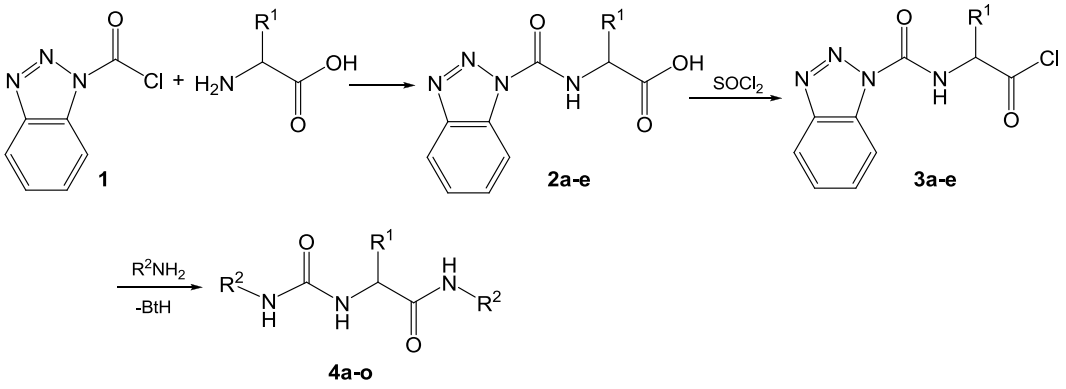


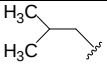
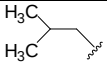
Shema 20. Sinteza klorida 1-benzotriazolkarboksilne kiseline (BtcCl, **1**)

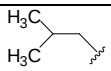
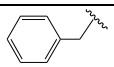
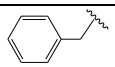
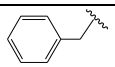
3.1.1. Sinteza aminokiselinskih derivata: ureidoamida 4a-o i *N*- i *O*-supstituiranih hidroksiurea 6a-l

Prvi korak u sintezi ureidoamida i hidroksiurea je sinteza *N*-(1-benzotriazolkarbonil)aminokiselina (*N*-Btc-aminokiselina) **2a-e**. Klorid 1-benzotriazol-karboksilne kiseline reagira s amino skupinom aminokiselina (L-alanin, L-valin, L-leucin, D-fenilglicin i L-fenilalanin) pri čemu nastaju *N*-Btc-aminokiseline (Butula *et al*, 1981a). Reakcija se provodi u bezvodnom dioksanu, na sobnoj temperaturi, uz množinski omjer reaktanata 1:2 (jedan mol aminokiseline služi kao akceptor klorovodika oslobođenog u reakciji, a drugi reagira s BtcCl). Nastali hidroklorid aminokiseline netopljiv je u dioksanu te se uklanja odsisavanjem, a uparavanjem matičnice dobije se sirovi produkt **2**.

N-(1-Benzotriazolkarbonil)aminokiseline **2a-e** prevedene su pomoću tionil-klorida ($SOCl_2$) u odgovarajuće kiselinske kloride **3a-e**, koji su upotrebljeni u daljnjim reakcijama aminolize bez čišćenja. Reakcijom s amino skupinom odgovarajućih aminoalkohola (2-aminoetanol, 3-aminopropanol i 5-aminopentanol) priređeni su ureidoamidi **4** (Shema 21) prema ranije opisanom postupku (Butula *et al*, 1981b). Množinski omjer aminoalkohola i klorida **3** iznosi 3:1 (dva mola aminoalkohola reagira s kloridom **3**, a treći služi kao akceptor klorovodika oslobođenog u reakciji). Reakcija se provodi na sobnoj temperaturi, gdje klorid reagira trenutno, dok je za supstituciju na benzotriazolskoj strani molekule potrebno više vremena. U zadanim uvjetima ne nastaje produkt klorida **3** s hidroksilnom skupinom aminoalkohola.



	4a	**4b**	**4c**	**4d**	**4e**	**4f**	**4g**	**4h**
R^1	CH_3	CH_3	CH_3					
R^2	$(CH_2)_2OH$	$(CH_2)_3OH$	$(CH_2)_5OH$	$(CH_2)_2OH$	$(CH_2)_3OH$	$(CH_2)_5OH$	$(CH_2)_2OH$	$(CH_2)_3OH$

	4i	**4j**	**4k**	**4l**	**4m**	**4n**	**4o**
R^1							
R^2	$(CH_2)_5OH$	$(CH_2)_2OH$	$(CH_2)_3OH$	$(CH_2)_5OH$	$(CH_2)_2OH$	$(CH_2)_3OH$	$(CH_2)_5OH$

Shema 21. Sinteza ureidoamida **4a-o**

Pripravljeni su sljedeći ureidoamidi:

1-(1-(2-hidroksietilkarbamoil)etil-3-(2-hidroksietil)urea (**4a**),

1-(1-(3-hidroksipropilkarbamoil)etil-3-(3-hidroksipropil)urea (**4b**),

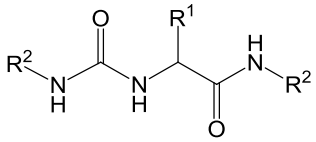
1-(1-(5-hidroksipentilkarbamoil)etil-3-(5-hidroksipentil)urea (**4c**),

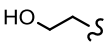
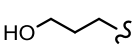
1-(1-(2-hidroksietilkarbamoil)-2-metilpropil-3-(2-hidroksietil)urea (**4d**),
1-(1-(3-hidroksipropilkarbamoil)-2-metilpropil-3-(3-hidroksipropil)urea (**4e**),
1-(1-(5-hidroksipentilkarbamoil)-2-metilpropil-3-(5-hidroksipentil)urea (**4f**),
1-(1-(2-hidroksietilkarbamoil)-3-metilbutil-3-(2-hidroksietil)urea (**4g**),
1-(1-(3-hidroksipropilkarbamoil)-3-metilbutil-3-(3-hidroksipropil)urea (**4h**),
1-(1-(5-hidroksipentilkarbamoil)-3-metilbutil-3-(5-hidroksipentil)urea (**4i**),
1-((2-hidroksietilkarbamoil)(fenil)metil)-3-(2-hidroksietil)urea (**4j**),
1-((3-hidroksipropilkarbamoil)(fenil)metil)-3-(3-hidroksipropil)urea (**4k**),
1-((3-hidroksipentilkarbamoil)(fenil)metil)-3-(5-hidroksipentil)urea (**4l**),
1-(1-(2-hidroksietilkarbamoil)-2-feniletil)-3-(2-hidroksietil)urea (**4m**),
1-(1-(3-hidroksipropilkarbamoil)-2-feniletil)-3-(3-hidroksipropil)urea (**4n**),
1-(1-(5-hidroksipentilkarbamoil)-2-feniletil)-3-(5-hidroksipentil)urea (**4o**).

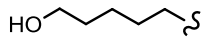
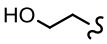
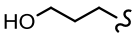
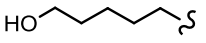
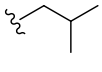
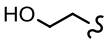
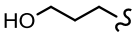
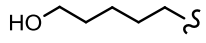
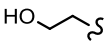
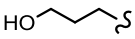
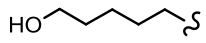
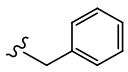
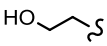
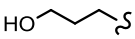
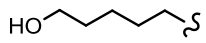
Sinteze novih spojeva i njihova čistoća praćeni su tankoslojnom kromatografijom, a strukture su potvrđene IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom.

IR spektri ureidoamida **4a-o** pokazuju karakteristične vrpce kod 3422–3092 (NH, OH), 1644–1625 (C=O, amid I) i 1559–1572 (C=O, amid II) cm^{-1} (Tablica 1). ^{1}H NMR spektri ispitivanih spojeva otkrivaju da se amidni NH nalazi u području 7,84–8,31 ppm, dok su karbamidni NH u području 5,88–6,78 ppm. Alkoholne skupine nalaze se u području 4,28–4,69 ppm (Tablica 2). Analizom ^{13}C NMR spektara utvrđeno je da se karbonil u položaju 1 (amid) nalazi u području 170,90–173,78 ppm, a karbonil u položaju 4 (urea) u području 157,49–158,45 ppm (Tablica 3).

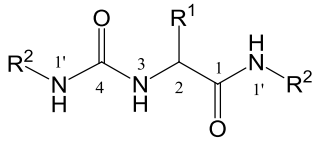
Tablica 1. Analitički i IR podaci za ureidoamide **4a-o**

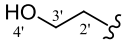


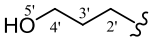
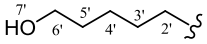
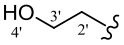
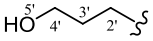
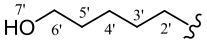
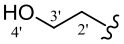
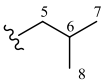
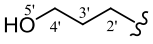
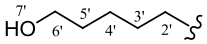
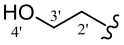
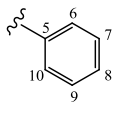
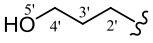
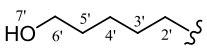
Spoj	**R^1**	**R^2**	**t_t (°C)**	**Mol. formula (M_r)**	**IR (KBr/film) ν_{max} (cm^{-1})**
4a	CH_3		150–154	$C_8H_{17}N_3O_4$ (219,24)	3422, 3274, 2969, 2937, 2879, 1644, 1570, 1056
4b			152–156	$C_{10}H_{21}N_3O_4$ (247,29)	3275, 2941, 2875, 1638, 1560, 1060

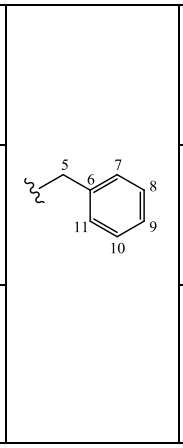
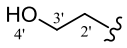
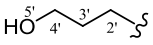
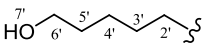
4c	CH_3		142–146	$C_{14}H_{29}N_3O_4$ (303,4)	3360, 3282, 3111, 2936, 2862, 1627, 1561, 1056
4d			194–198	$C_{10}H_{21}N_3O_4$ (247,29)	3280, 3102, 2969, 2874, 1626, 1571, 1063
4e			178–181	$C_{12}H_{25}N_3O_4$ (275,34)	3350, 3279, 3102, 2963, 2874, 1630, 1566, 1234, 1078, 1044
4f			159–162	$C_{16}H_{33}N_3O_4$ (331,45)	3349, 3278, 3107, 2935, 2861, 1628, 1572, 1236, 1059
4g			153–161	$C_{11}H_{23}N_3O_4$ (261,32)	3290, 3098, 2954, 2928, 2872, 1625, 1569, 1059
4h			109–111	$C_{13}H_{27}N_3O_4$ (289,37)	3308, 3098, 2957, 2872, 1630, 1572, 1252, 1073
4i			83–86	$C_{17}H_{35}N_3O_4$ (345,48)	3349, 3280, 3100, 2937, 2862, 1626, 1562, 1262, 1059
4j			206–208	$C_{13}H_{19}N_3O_4$ (281,31)	3291, 3092, 2934, 2878, 1626, 1571, 1226, 1057
4k			182–185	$C_{15}H_{23}N_3O_4$ (309,36)	3289, 3092, 2942, 2883, 1626, 1561, 1059
4l			178–180	$C_{19}H_{31}N_3O_4$ (365,47)	3362, 3286, 3093, 2937, 2861, 1628, 1564, 1352, 1061
4m			140–143	$C_{14}H_{21}N_3O_4$ (295,33)	3310, 3105, 2937, 2876, 1626, 1569, 1496, 1231, 1056
4n			110–113	$C_{16}H_{25}N_3O_4$ (323,39)	3280, 3106, 2942, 2878, 1628, 1564, 1235, 1050
4o			89–93	$C_{20}H_{33}N_3O_4$ (379,49)	3282, 3106, 2931, 2859, 1626, 1559, 1236, 1060

Tablica 2. ^{1}H NMR spektroskopski podaci za ureidoamide **4a-o**

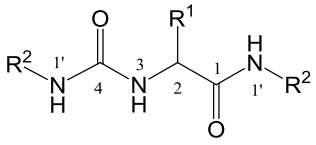


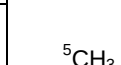
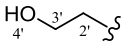
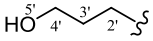
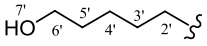
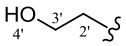
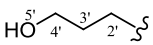
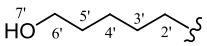
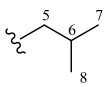
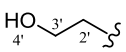
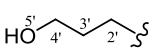
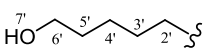
Spoj	**R^1**	**R^2**	**^{1}H NMR (DMSO-d_6, δ ppm)**
4a	$^5CH_3$		7,87 (t, 1H, 1'(amid), *J* = 5,4 Hz), 6,21 (d, 1H, 3, *J* = 7,9 Hz), 6,14 (t, 1H, 1'(urea), *J* = 5,5 Hz), 4,69, 4,67 (2t, 2H, 4', *J* = 5,6 Hz, *J* = 5,2 Hz), 4,16–4,06 (m, 1H, 2), 3,38–3,32 (m, 4H, 3'), 3,13–3,00 (m, 4H, 2'), 1,12 (d, 3H, 5, *J* = 7,4 Hz)

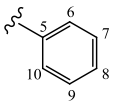
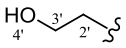
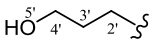
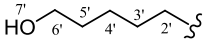
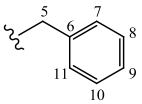
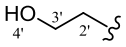
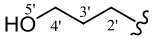
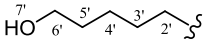
4b	$^{5}CH_3$		7,85 (t, 1H, 1'(amid), *J* = 5,5 Hz), 6,05–6,02 (d, t, 2H, 1'(urea), 3), 4,43, 4,42 (2t, 2H, 5', *J* = 4,8 Hz, *J* = 5,1 Hz), 4,14–4,05 (m, 1H, 2), 3,39 (q, 4H, 4', *J* = 6,1 Hz), 3,12–3,00 (m, 4H, 2'), 1,58–1,44 (m, 4H, 3'), 1,12 (d, 3H, 5, *J* = 6,8 Hz)
4c			7,84 (t, 1H, 1'(amid), *J* = 5,3 Hz), 6,03 (t, 1H, 1'(urea), *J* = 5,5 Hz), 5,96 (d, 1H, 3, *J* = 7,8 Hz), 4,34 (t, 2H, 7', *J* = 5,1 Hz), 4,15–4,05 (m, 1H, 2), 3,37 (q, 4H, 6', *J* = 5,8 Hz), 3,05–2,92 (m, 4H, 2'), 1,45–1,29 (m, 12H, 3'–5'), 1,12 (d, 3H, 5)
4d			7,87 (t, 1H, 1'(amid), *J* = 5,4 Hz), 6,16 (t, 1H, 1'(urea), *J* = 5,5 Hz), 6,11 (d, 1H, 3, *J* = 9,1 Hz), 4,65, 4,64 (2t, 2H, 4', *J* = 4,8 Hz, *J* = 5,1 Hz), 3,97 (dd, 1H, 2, *J* = 6,0 Hz, *J* = 2,9 Hz), 3,45–3,33 (m, 4H, 3'), 3,18–3,03 (m, 4H, 2'), 1,91–1,80 (m, 1H, 5), 0,82 (d, 3H, 6, *J* = 6,8 Hz), 0,78 (d, 3H, 7, *J* = 6,8 Hz)
4e			7,90 (t, 1H, 1'(amid), *J* = 5,6 Hz), 6,06 (t, 1H, 1'(urea), *J* = 5,7 Hz), 5,96 (d, 1H, 3, *J* = 9,1 Hz), 4,42, 4,41 (2t, 2H, 5', *J* = 5,4 Hz), 3,95 (dd, 1H, 2, *J* = 6,1 Hz, *J* = 3,0 Hz), 3,39 (q, 4H, 4', *J*=6,0 Hz), 3,19–3,00 (m, 4H, 2'), 1,89–1,78 (m, 1H, 5), 1,58–1,45 (m, 4H, 3'), 0,82 (d, 3H, 6, *J* = 6,8 Hz), 0,79 (d, 3H, 7, *J* = 7,0 Hz)
4f			7,89 (t, 1H, 1'(amid), *J* = 5,5 Hz), 6,05 (t, 1H, 1'(urea), *J* = 5,4 Hz), 5,90 (d, 1H, 3, *J* = 9,0 Hz), 4,35–4,32 (m, 2H, 7'), 3,95 (dd, 1H, 2, *J* = 6,2 Hz, *J* = 2,7 Hz), 3,38–3,35 (m, 4H, 6'), 3,12–2,93 (m, 4H, 2'), 1,88–1,77 (m, 1H, 5), 1,44–1,25 (m, 12H, 3'–5'), 0,81 (d, 3H, 6, *J* = 6,8 Hz), 0,78 (d, 3H, 7, *J* = 6,9 Hz)
4g			7,87 (t, 1H, 1'(amid), *J* = 5,3 Hz), 6,11 (d, 1H, 3, *J* = 8,9 Hz), 6,04 (t, 1H, 1'(urea), *J* = 5,4 Hz), 4,65–4,64 (m, 2H, 4'), 4,15–4,07 (m, 1H, 2), 3,40–3,34 (m, 4H, 3'), 3,13–3,01 (m, 4H, 2'), 1,60–1,52 (m, 1H, 6), 1,42–1,27 (m, 2H, 5), 0,86 (2d, 6H, 7, 8, *J* = 6,2 Hz, *J* = 5,4 Hz)
4h			7,89 (t, 1H, 1'(amid), *J* = 5,5 Hz), 5,97–5,94 (m, 2H, 3, 1'(urea)), 4,42, 4,40 (2t, 2H, 5', *J* = 5,3 Hz), 4,13–4,05 (m, 1H, 2), 3,42–3,35 (m, 4H, 4'), 3,15–2,99 (m, 4H, 2'), 1,59–1,44 (m, 5H, 6, 3'), 1,39–1,30 (m, 2H, 5), 0,87 (d, 3H, 7, *J* = 6,2 Hz), 0,85 (d, 3H, 8, *J* = 6,2 Hz)
4i			7,88 (t, 1H, 1'(amid), *J* = 5,4 Hz), 5,94 (t, 1H, 1'(urea), *J* = 5,7 Hz), 5,88 (d, 1H, 3, *J* = 9,0 Hz), 4,33 (t, 2H, 7', *J* = 5,1 Hz), 4,13–4,06 (m, 1H, 2), 3,39–3,33 (m, 4H, 6'), 3,08–2,92 (m, 4H, 2'), 1,58–1,22 (m, 15H, 6, 5, 3'–5'), 1,39–1,30 (m, 2H, 5), 0,87 (d, 3H, 7, *J* = 6,1 Hz), 0,85 (d, 3H, 8, *J* = 6,1 Hz)
4j			8,31 (t, 1H, 1'(amid), *J* = 5,5 Hz), 7,37–7,21 (m, 5H, arom., 6–10), 6,78 (d, 1H, 3, *J* = 8,4 Hz), 6,29 (t, 1H, 1'(urea), *J* = 5,5 Hz), 5,31 (d, 1H, 2, *J* = 8,4 Hz), 4,66, 4,64 (2t, 2H, 4', *J* = 5,3 Hz, *J* = 5,1 Hz), 3,40–3,34 (m, 4H, 3'), 3,23–2,97 (m, 4H, 2')
4k			8,27 (t, 1H, 1'(amid), *J* = 5,5 Hz), 7,36–7,22 (m, 5H, arom., 6–10), 6,65 (d, 1H, 3, *J* = 8,3 Hz), 6,21 (t, 1H, 1'(urea), *J* = 5,5 Hz), 5,26 (d, 1H, 2, *J* = 8,5 Hz), 4,41, 4,39 (2t, 2H, 5', *J* = 5,2 Hz), 3,42–3,32 (m, 4H, 4'), 3,12–2,99 (m, 4H, 2'), 1,56–1,44 (m, 4H, 3')
4l			8,26 (t, 1H, 1'(amid), *J* = 5,5 Hz), 7,36–7,21 (m, 5H, arom., 6–10), 6,60 (d, 1H, 3, *J* = 8,4 Hz), 6,21 (t, 1H, 1'(urea), *J* = 5,5 Hz), 5,26 (d, 1H, 2, *J* = 8,4 Hz), 4,35, 4,31 (2t, 2H, 7', *J* = 5,3 Hz), 3,40–3,30 (m, 4H, 6'), 3,05–2,92 (m, 4H, 2'), 1,44–1,16 (m, 12H, 3'–5')

4m			7,90 (t, 1H, 1'(amid), *J* = 5,1 Hz), 7,28–7,17 (m, 5H, arom., 6–10), 6,16 (d, 1H, 3, *J* = 8,6 Hz), 6,10 (t, 1H, 1'(urea), *J* = 5,1 Hz), 4,64–4,59 (m, 2H, 4'), 4,37–4,30 (m, 1H, 2), 3,33–3,29 (m, 4H, 3'), 3,12–2,67 (m, 6H, 2', 5)
4n			7,88 (t, 1H, 1'(amid)', *J* = 4,6 Hz), 7,27–7,14 (m, 5H, arom., 6–10), 6,04–6,02 (d, t, 2H, 3, 1'(urea)), 4,42–4,28 (m, 3H, 2, 5'), 3,39–3,34 (m, 4H, 4'), 3,12–2,69 (m, 6H, 2', 5), 1,50–1,43 (m, 4H, 3')
4o			7,86 (t, 1H, 1'(amid), *J* = 5,5 Hz), 7,27–7,14 (m, 5H, arom., 6–10), 6,01 (t, 1H, 1'(urea), *J* = 5,5 Hz), 5,96 (d, 1H, 3, *J* = 8,7 Hz), 4,35–4,28 (m, 3H, 2, 7'), 3,37–3,33 (m, 4H, 6'), 3,07–2,69 (m, 6H, 2', 5), 1,43–1,16 (m, 12H, 3'–5')

Tablica 3. ^{13}C NMR spektroskopski podaci za ureidoamide **4a-o**



Spoj	**R^1**	**R^2**	**^{13}C NMR (DMSO-d_6, δ ppm)**
4a			173,78 (1), 157,98 (4), 61,19, 60,21 (3'), 49,07 (2), 42,46, 41,86 (2'), 20,11 (5)
4b			173,63 (1), 157,97 (4), 58,83, 58,82 (4'), 49,02 (2), 36,70, 36,12 (2'), 33,66, 32,79 (3'), 20,23 (5)
4c			173,44 (1), 157,77 (4), 61,13, 61,08 (6'), 48,95 (2), 39,63, 38,89 (2'), 32,72, 32,63 (5'), 30,35, 29,39 (3'), 23,39, 23,30 (4'), 20,36 (5)
4d			172,60 (1), 158,44 (4), 61,25, 60,28 (3'), 58,34 (2), 42,49, 41,76 (2'), 31,59 (5), 19,73, 18,25 (6, 7)
4e			172,40 (1), 158,45 (4), 58,89, 58,79 (4'), 58,28 (2), 36,68, 36,07 (2'), 33,71, 32,86 (3'), 31,72 (5), 19,71, 18,35 (6, 7)
4f			172,23 (1), 158,25 (4), 61,13, 61,08 (6'), 58,21 (2), 39,67, 38,83 (2'), 32,72, 32,62 (5'), 31,78 (5), 30,35, 29,41 (3'), 23,39, 23,35 (4'), 19,70, 18,33 (6,7)
4g			173,69 (1), 158,18 (4), 61,21, 60,24 (3'), 51,95 (2), 42,84, 42,49 (2'), 41,83 (5), 24,69 (6), 23,51, 22,39 (7, 8)
4h			173,50 (1), 158,19 (4), 58,90, 58,85 (4'), 51,95 (2), 42,92 (5), 36,73, 36,11 (2'), 33,68, 32,81 (3'), 24,74 (6), 23,43, 22,49 (7, 8)
4i			173,29 (1), 157,98 (4), 61,13, 61,09 (6'), 51,90 (2), 43,03 (5), 39,70, 38,87 (2'), 32,73, 32,64 (5'), 30,35, 29,37 (3'), 24,74 (6),

			23,41, 22,57 (7, 8), 23,38, 23,31 (4')
4j			171,25 (1), 157,63 (4), 141,05 (5), 128,61, 127,10 (6, 7, 9 10), 127,57 (8), 61,18, 60,11 (3'), 56,93 (2), 42,46, 42,01 (2')
4k			171,06 (1), 157,67 (4), 141,10 (5), 128,63, 126,96 (6, 7, 9, 10), 127,59 (8), 58,79, 58,72 (4'), 56,98 (2), 39,15, 36,69 (2'), 36,33, 36,28 (3')
4l			170,90 (1), 157,49 (4), 141,21 (5), 128,59, 126,93 (6, 7, 9, 10), 127,54 (8), 61,13, 61,06 (6'), 56,94 (2), 39,63, 39,04 (2'), 32,71, 32,58 (5'), 30,31, 29,24 (3'), 23,38, 23,25 (4')
4m			172,51 (1), 158,00 (4), 138,41 (6), 129,71, 128,43 (7, 8, 10, 11), 126,58 (9), 61,16, 60,18 (3'), 54,80 (2), 42,42, 41,81 (2'), 39,42 (5)
4n			172,28 (1), 158,00 (4), 138,35 (5), 129,68, 128,43 (7, 8, 10, 11), 126,58 (9), 58,93 (4'), 54,79 (2), 39,46 (5), 36,68, 36,14 (2'), 33,61, 32,72 (3')
4o			172,06 (1), 157,79 (4), 138,34 (5), 129,70, 128,40 (7, 8, 10, 11), 126,55 (9), 61,13, 61,07 (6'), 54,72 (2), 39,62, 39,55 (2'), 38,91 (5), 32,72, 32,67 (5'), 30,30, 29,33 (3'), 23,36, 23,29 (4')

Ureidoamidi **4b**,**i**,**j**,**m** poslužili su kao alkoholi u sintezi estera ketoprofena **12a-d**.

Reakcijom klorida *N*-(1-benzotriazolkarbonil)aminokiseline **3c**-**e** s različitim aminima (cikloheksilamin, cikloheksanmetilamin i benzhidrilamin) sintetizirani su amidi *N*-(1-benzotriazolkarbonil)aminokiselina **5a-f** prema ranije opisanom postupku (Butula *et al*, 1981b), a opisana je i sinteza i potpuna karakterizacija (tališta, elementarne analize, IR, ^{1}H i ^{13}C NMR) tih i srodnih spojeva (Opačić *et al*, 2005).

Pripravljeni su sljedeći amidi:

N-(1-benzotriazolkarbonil)-L-leucin benzhidrilamid (**5a**),

N-(1-benzotriazolkarbonil)-D-fenilglicin cikloheksanmetilamid (**5b**),

N-(1-benzotriazolkarbonil)-D-fenilglicin benzhidrilamid (**5c**),

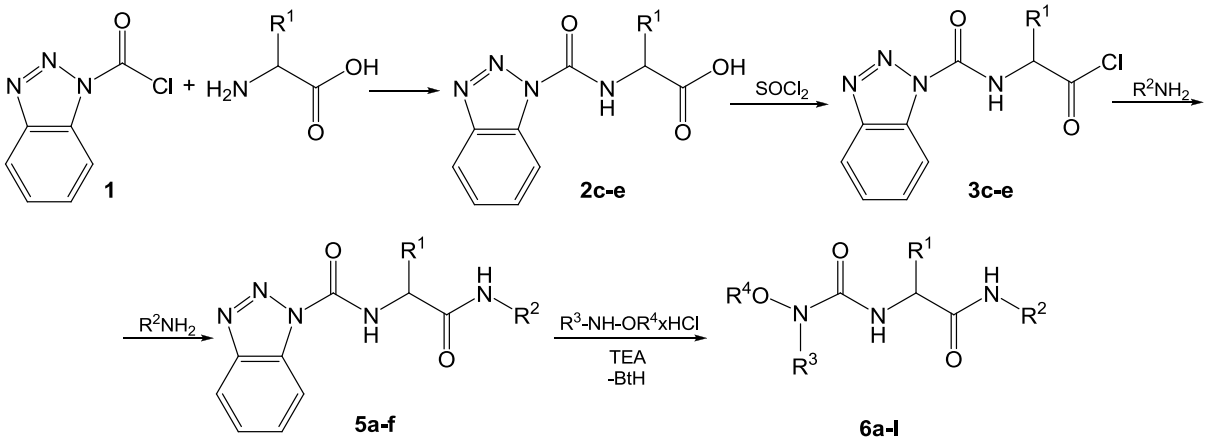
N-(1-benzotriazolkarbonil)-L-fenilalanin cikloheksilamid (**5d**),

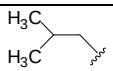
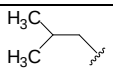
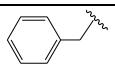
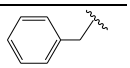
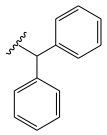
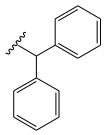
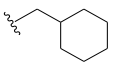
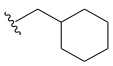
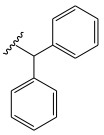
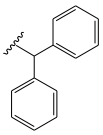
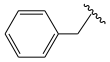
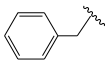
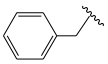
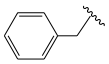
N-(1-benzotriazolkarbonil)-L-fenilalanin cikloheksanmetilamid (**5e**),

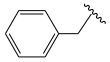
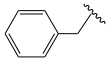
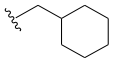
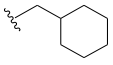
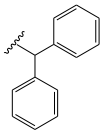
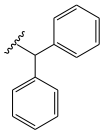
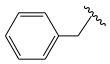
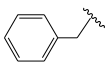
N-(1-benzotriazolkarbonil)-L-fenilalanin benzhidrilamid (**5f**).

Novi derivati hidroksiuree **6a-l** (Shema 22) sintetizirani su aminolizom amida *N*-(1-benzotriazolkarbonil)aminokiselina **5a-f** s *O*-benzilhidroksilaminom ili *N*-metilhidroksil-

aminom uz TEA koji služi kao akceptor klorovodika oslobođenog iz hidroklorida hidroksilamina prema ranije opisanom postupku (Opačić *et al*, 2005).



	6a	6b	6c	6d	6e	6f	6g	6h
R^1								
R^2								
R^3	H	$-CH_3$	H	$-CH_3$	H	$-CH_3$	H	$-CH_3$
R^4		H		H		H		H

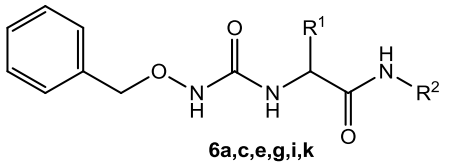
	6i	6j	6k	6l
R^1				
R^2				
R^3	H	$-CH_3$	H	$-CH_3$
R^4		H		H

Shema 22. Sinteza *N*- i *O*-supstituiranih hidroksiurea **6a-l**

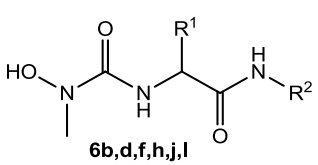
Pripravljene su sljedeće *N*- i *O*-supstituirane hidroksiuree:
N-benzhidrilamid-2-(*N*'-benziloksiureido)-l-4-metilpentanske kiseline (**6a**),
N-benzhidrilamid-2-(*N*'-metil-*N*'-hidroksiureido)-l-4-metilpentanske kiseline (**6b**),
N-cikloheksanmetilamid-2-(*N*'-benziloksiureido)-d-2-feniletanske kiseline (**6c**),
N-cikloheksanmetilamid-2-(*N*'-metil-*N*'-hidroksiureido)-d-2-feniletanske kiseline (**6d**),
N-benzhidrilamid-2-(*N*'-benziloksiureido)-d-2-feniletanske kiseline (**6e**),
N-benzhidrilamid-2-(*N*'-metil-*N*'-hidroksiureido)-d-2-feniletanske kiseline (**6f**),
N-cikloheksilamid-2-(*N*'-benziloksiureido)-l-3-fenilpropanske kiseline (**6g**),
N-cikloheksilamid-2-(*N*'-metil-*N*'-hidroksiureido)-l-3-fenilpropanske kiseline (**6h**),
N-cikolheksanmetilamid-2-(*N*'-benziloksiureido)-l-3-fenilpropanske kiseline (**6i**),
N-cikloheksanmetilamid-2-(*N*'-metil-*N*'-hidroksiureido)-l-3-fenilpropanske kiseline (**6j**),
N-benzhidrilamid-2-(*N*'-benziloksiureido)-l-3-fenilpropanske kiseline (**6k**),
N-benzhidrilamid-2-(*N*'-metil-*N*'-hidroksiureido)-l-3-fenilpropanske kiseline (**6l**).

IR spektri hidroksiurea **6a-l** pokazuju karakteristične vrpce kod 3439–3030 (NH, OH), 1627–1667 (C=O, amid I) i 1520–1584 (C=O, amid II) cm^{-1} (Tablica 4). ^{1}H NMR spektri ispitivanih spojeva otkrivaju da se jedan karbamidni NH (3) nalazi u području 6,45–7,07 ppm, dok je drugi karbamidni NH (1', prisutan samo u *O*-benzilnim derivatima) u području 9,17–9,40 ppm. OH (prisutan samo u *N*-metilnim derivatima) se nalaze u području 9,48–9,85 ppm. Zanimljiva je pojava cijepanja signala metilenskog mosta na dublet dubleta s velikom konstantom povratne sprege (J = 10,94–11,6 Hz) u *O*-benzilu koja je uočena u nekoliko derivata; **6a,c,e** dok je u **6g,i,k** taj signal prisutan u obliku očekivanog singleta (Tablica 5). Analizom ^{13}C NMR spektara utvrđeno je da se karbonil u položaju 1 (amid) nalazi u području 169,08–171,81 ppm, a karbonil u položaju 4 (urea) u području 158,12–160,71 ppm. Kemijski pomaci protona u ^{1}H, te atoma ugljika u ^{13}C NMR spektrima u skladu su s predloženim strukturama novih spojeva (Tablica 6).

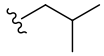
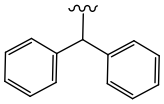
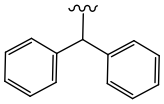
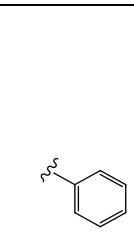
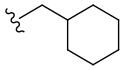
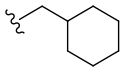
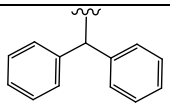
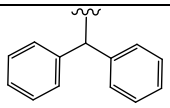
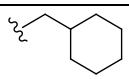
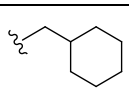
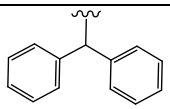
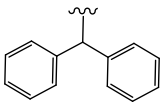
Tablica 4. Analitički i IR podaci za hidroksiuree **6a**-**l**



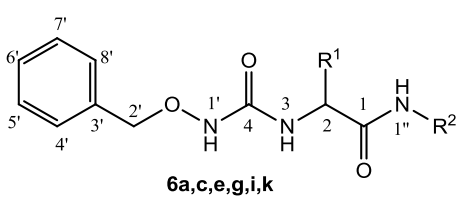
6a,c,e,g,i,k



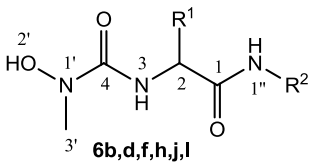
6b,d,f,h,j,l

Spoj	R^1	R^2	t_t (°C)	Mol. formula (M_r)	IR (KBr) ν_{max} (cm^{-1})
6a			100–103	$C_{27}H_{31}N_3O_3$ (445,55)	3308, 3061, 3030, 2957, 2930, 2872, 1645, 1525, 1496, 1453, 1336, 1240, 1212, 1078, 1028, 902, 752, 698, 607
6b			219–222 (raspad)	$C_{21}H_{27}N_3O_3$ (369,46)	3407, 3309, 3162, 3031, 2964, 2930, 2900, 2872, 1648, 1548, 1515, 1496, 1471, 1450, 1437, 1368, 1296, 1185, 1173, 1120, 761, 749, 701
6c			148–150	$C_{23}H_{29}N_3O_3$ (395,49)	3310, 3091, 3063, 3032, 2926, 2854, 1639, 1533, 1450, 1355, 1221, 1154, 985, 931, 907, 747, 697
6d			123–126	$C_{17}H_{25}N_3O_3$ (319,40)	3425, 3310, 2925, 2853, 1650, 1532, 1450, 1367, 1184, 1120, 698, 668
6e			161–165	$C_{29}H_{27}N_3O_3$ (465,54)	3308, 3062, 3031, 2934, 1667, 1642, 1536, 1496, 1453, 1366, 1231, 1213, 1031, 934, 743, 700, 640, 614
6f			165–168 (raspad)	$C_{23}H_{23}N_3O_3$ (389,45)	3439, 3376, 3210, 3064, 3031, 2924, 2892, 1637, 1584, 1520, 1496, 1450, 1367, 1177, 1120, 1030, 748, 697, 636
6g			129–131	$C_{23}H_{29}N_3O_3$ (395,49)	3296, 3206, 3064, 3031, 2931, 2854, 1639, 1533, 1498, 1453, 1364, 1250, 1075, 952, 814, 750, 698
6h			153–155	$C_{17}H_{25}N_3O_3$ (319,40)	3390, 3289, 3089, 2935, 2854, 1648, 1627, 1550, 1452, 1390, 1182, 751, 702
6i			130–131	$C_{24}H_{31}N_3O_3$ (409,52)	3313, 3214, 3064, 3031, 2921, 2852, 1661, 1640, 1542, 1496, 1449, 1363, 1276, 1242, 1072, 948, 813, 748, 698, 632, 616
6j			157–159	$C_{18}H_{27}N_3O_3$ (333,43)	3371, 3329, 3194, 2924, 2853, 1658, 1642, 1580, 1528, 1495, 1445, 1190, 1123, 746, 700, 669
6k			161–164	$C_{30}H_{29}N_3O_3$ (479,57)	3288, 3062, 3030, 1649, 1535, 1495, 1454, 1229, 1030, 910, 750, 698
6l			152–154 (raspad)	$C_{24}H_{25}N_3O_3$ (403,47)	3432, 3347, 3062, 3030, 2867, 1664, 1629, 1536, 1496, 1450, 1384, 1230, 1179, 1117, 1030, 744, 700

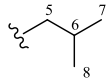
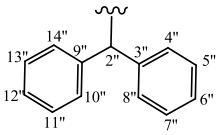
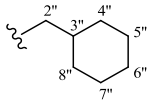
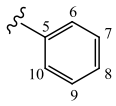
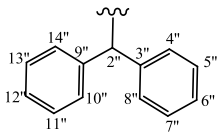
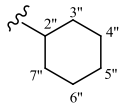
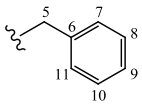
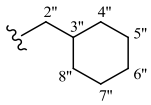
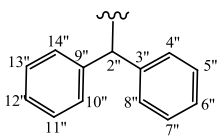
Tablica 5. ^{1}H NMR za hidroksiuree **6a-l**



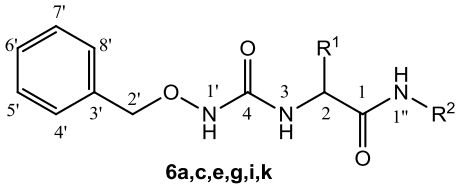
6a,c,e,g,i,k



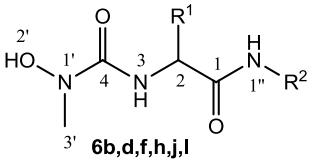
6b,d,f,h,j,l

Spoj	R^1	R^2	^{1}H NMR (DMSO-d_6, δ ppm)
6a			9,18 (s, 1H, 1'), 8,82 (d, 1H, 1", *J* = 8,51 Hz), 7,33–7,16 (m, 15H, arom.), 6,45 (d, 1H, 3, *J* = 8,92 Hz), 6,03 (d, 1H, 2", *J* = 8,51 Hz), 4,64 (dd, 2H, 2', $J_{vic\ H,H}$ = 11,6 Hz), 4,32–4,28 (m, 1H, 2), 1,43–1,32 (m, 3H, 5, 6), 0,79 (2d, 6H, 7, 8, *J* = 6,43 Hz)
6b			9,52 (s, 1H, 2'), 8,90 (d, 1H, 1", *J* = 8,47 Hz), 7,38–7,18 (m, 10H, arom.), 6,68 (d, 1H, 3, *J* = 8,97 Hz), 6,10 (d, 1H, 2", *J* = 8,47 Hz), 4,38–4,32 (m, 1H, 2), 2,95 (s, 3H, 3'), 1,58–1,39 (m, 3H, 5, 6), 0,85 (2d, 6H, 7, 8, *J* = 3,24 Hz)
6c			9,40 (s, 1H, 1'), 8,29 (t, 1H, 1", *J* = 5,57 Hz), 7,43–7,22 (m, 10H, arom.), 6,91 (d, 1H, 3, *J* = 7,98 Hz), 5,28 (d, 1H, 2, *J* = 7,98 Hz), 4,76 (dd, 2H, 2', $J_{vic\ H,H}$ = 11,26 Hz), 2,93–2,82 (m, 2H, 2"), 1,61–0,83 (m, 11H, 3"–8")
6d			9,85 (bs, 1H, 2'), 8,33 (t, 1H, 1", *J* = 5,76 Hz), 7,39–7,23 (m, 5H, arom.), 7,07 (d, 1H, 3, *J* = 8,16 Hz), 5,28 (d, 1H, 2, *J* = 8,16 Hz), 2,95 (s, 3H, 3'), 2,92–2,87 (m, 2H, 2"), 1,60–0,77 (m, 11H, 3"–8")
6e			9,33 (s, 1H, 1'), 9,19 (d, 1H, 1", *J* = 8,28 Hz), 7,35–6,98 (m, 20H, arom.), 6,85 (d, 1H, 3, *J* = 7,96 Hz), 6,01 (d, 1H, 2", *J* = 7,96 Hz), 5,43 (d, 1H, 2, *J* = 7,96 Hz), 4,68 (dd, 2H, 2', $J_{vic\ H,H}$ = 10,94 Hz)
6f			9,71 (s, 1H, 2'), 9,31 (d, 1H, 1", *J* = 8,40 Hz), 7,45–7,04 (m, 16H, arom., 3), 6,09 (d, 1H, 2", *J* = 8,19 Hz), 5,53 (d, 1H, 2, *J* = 8,19 Hz), 2,96 (s, 3H, 3')
6g			9,24 (s, 1H, 1'), 7,88 (d, 1H, 1", *J* = 7,66 Hz), 7,35–7,13 (m, 10H, arom.), 6,53 (d, 1H, 3, *J* = 8,61 Hz), 4,64 (s, 1H, 2'), 4,41 (q, 1H, 2, *J* = 7,48 Hz), 3,52–3,49 (m, 1H, 2"), 2,96–2,88 (d, 2H, 5), 1,73–1,03 (m, 10H, 3"–7")
6h			9,50 (s, 1H, 2'), 7,83 (d, 1H, 1", *J* = 7,84 Hz), 7,27–7,15 (m, 5H, arom.), 6,59 (d, 1H, 3, *J* = 8,56 Hz), 4,36 (q, 1H, 2, *J* = 6,82 Hz, 8,27 Hz), 3,52–3,43 (m, 1H, 2"), 2,91 (s, 3H, 3'), 2,88 (d, 2H, 5, *J* = 6,97 Hz), 1,71–1,02 (m, 10H, 3"–7")
6i			9,24 (s, 1H, 1'), 7,96 (t, 1H, 1", *J* = 5,57 Hz), 7,39–7,19 (m, 10H, arom.), 6,56 (d, 1H, 3, *J* = 8,44 Hz), 4,65 (s, 1H, 2'), 4,43 (q, 1H, 2, *J* = 8,12, 5,89 Hz), 2,99–2,82 (m, 4H, 2", 5), 1,66–0,81 (m, 11H, 3"–8")
6j			9,48 (s, 1H, 2'), 7,93 (t, 1H, 1", *J* = 5,21 Hz), 7,29–7,16 (m, 5H, arom.), 6,63 (d, 1H, 3, *J* = 8,54 Hz), 4,38 (q, 1H, 2, *J* = 7,32 Hz), 2,97–2,75 (m, 7H, 5, 2"), 2,91 (s, 3H, 3'), 1,69–0,76 (m, 11H, 3"–8")
6k			9,17 (s, 1H, 1'), 8,89 (d, 1H, 1", *J* = 8,47 Hz), 7,32–7,11 (m, 20H, arom.), 6,54 (d, 1H, 3, *J* = 8,74 Hz), 6,03 (d, 1H, 2", *J* = 8,34 Hz), 4,54 (q+s, 3H, 2, 2'), 2,93–2,84 (m, 2H, 5)
6l			9,52 (s, 1H, 2'), 8,96 (d, 1H, 1", *J* = 8,32 Hz), 7,37–7,15 (m, 15H, arom.), 6,69 (d, 1H, 3, *J* = 8,32 Hz), 6,09 (d, 1H, 2", *J* = 8,32 Hz), 4,60 (q, 1H, 2, *J* = 6,93 Hz), 2,95–2,91 (m, 2H, 5), 2,91 (s, 3H, 3')

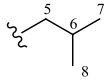
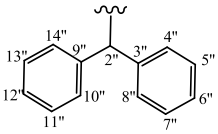
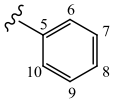
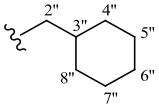
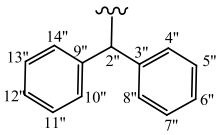
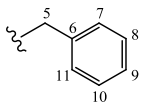
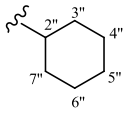
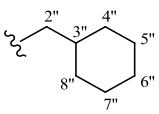
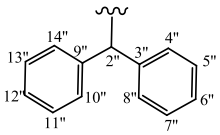
Tablica 6. ^{13}C NMR za hidroksiuree **6a-l**

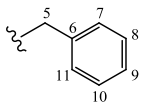
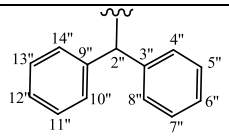


6a,c,e,g,i,k



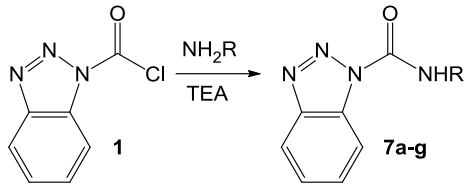
6b,d,f,h,j,l

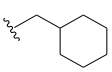
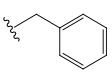
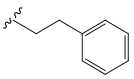
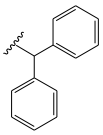
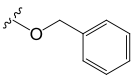
Spoj	R^1	R^2	^{13}C NMR (DMSO-d_6, δ ppm)
6a			171,66 (1), 158,95 (4), 142,25, 142,21 (3", 9"), 136,26 (3'), 128,73, 128,35, 128,30, 128,26, 127,39, 127,09 (4', 5', 7', 8', 4", 5", 7", 8", 10", 11", 13", 14"), 128,09, 126,94, 126,90 (7', 6", 12"), 77,40 (2'), 55,84 (2"), 51,16 (2), 41,67 (5), 24,17, 23,04, 21,72 (6–8)
6b			171,81 (1), 160,34 (4), 142,30 (3", 9"), 128,39, 128,35, 127,41, 127,10 (4", 5", 7", 8", 10", 11", 13", 14"), 127,01, 126,97 (6", 12"), 55,84 (2"), 51,85 (2), 42,03 (5), 38,59 (3'), 24,24, 23,05, 21,84 (6–8)
6c			169,68 (1), 158,24 (4), 139,79 (5), 136,09 (3'), 129,00, 128,42, 128,20, 126,37 (6, 7, 9, 10, 4', 5', 7', 8'), 128,29, 127,36 (8, 7'), 77,54 (2'), 55,76 (2), 44,88 (2"), 37,35 (3"), 30,19, 30,14, 25,32 (4", 5", 7", 8"), 25,93 (6")
6d			170,32 (1), 160,05 (4), 140,51 (5), 128,69, 126,89 (6, 7, 9, 10), 127,84 (8), 56,86 (2), 45,34 (2"), 38,76 (3'), 37,81 (3"), 30,66, 30,62, 25,78 (4", 5", 7", 8"), 26,39 (6")
6e			169,08 (1), 158,25 (4), 141,90, 141,77 (3", 9"), 139,34 (5), 136,05 (3'), 128,94, 128,42, 128,38, 128,23, 127,51, 126,79, 126,46 (6, 7, 9, 10, 4', 5', 7', 8', 4", 5", 7", 8", 10", 11", 13", 14"), 77,52 (2'), 56,02, 55,63 (2, 2")
6f			169,72 (1), 160,03 (4), 142,47, 142,25 (3", 9"), 140,11 (5), 128,93, 128,77, 128,74, 127,97, 127,31, 127,01 (6, 7, 9, 10, 4", 5", 7", 8", 10", 11", 13", 14"), 27,97, 127,66, 127,43 (8, 6", 12"), 56,68, 56,51 (2, 2"), 38,78 (3')
6g			170,36 (1), 159,14 (4), 137,93 (6), 136,59 (3'), 129,82, 129,17, 128,36 (7, 8, 10, 11, 4', 5', 7', 8'), 128,60, 126,63 (9, 7'), 77,84 (2'), 54,00 (2), 47,96 (2"), 39,01 (5), 32,74, 32,73, 24,90, 24,85 (3", 4", 6", 7"), 25,65 (5")
6h			170,38 (1), 160,61 (4), 137,91 (6), 129,77, 128,43 (8, 7, 10, 11), 126,68 (9), 54,62 (2), 47,86 (2"), 39,32 (5), 38,99 (3'), 32,70, 24,92, 24,87 (3", 4", 6", 7"), 25,64 (5")
6i			170,38 (1), 158,12 (4), 136,98 (3'), 135,56 (6), 128,68, 128,08, 127,69, 127,43 (7, 8, 10, 11, 4', 5', 7', 8'), 127,52, 125,67 (9, 7'), 76,78 (2'), 53,22 (2), 44,24 (2"), 37,81 (5), 36,74 (3"), 29,72, 24,82 (4", 5", 7", 8"), 25,42 (6")
6j			171,48 (1), 160,64 (4), 138,03 (6), 129,68, 128,47 (7, 8, 10, 11), 126,67 (9), 54,88 (2), 45,26 (2"), 39,14 (5), 38,98 (3'), 37,77 (3"), 30,76, 25,87 (4", 5", 7", 8"), 26,46 (6")
6k			170,55 (1), 158,75 (4), 142,15, 142,14 (3", 9"), 137,41 (6), 136,10 (3'), 129,31, 128,62, 128,34, 128,25, 128,00, 127,39, 127,14 (7, 8, 10, 11, 4", 5", 7", 8", 10", 11", 13", 14"), 128,07, 127,00, 126,91, 126,28 (9, 6", 12"), 77,34 (5), 55,96, 53,75 (2, 2"), 38,16 (3')

6l			171,10 (1), 160,71 (4), 142,70, 142,63 (3", 9"), 137,91 (6), 129,78, 128,84, 128,78, 128,51, 127,89, 127,61 (7, 8, 10, 11, 4", 5", 7", 8", 10", 11", 13", 14"), 127,51, 127,39, 126,73 (9, 6", 12"), 56,37, 54,87 (2, 2"), 38,98 (3')

3.1.2. Sinteza 1-karbamoilbenzotriazola (amida 1-benzotriazolkarboksilne kiseline) 7a-g

Klorid **1** u reakciji s aminima (ciklopentilamin, cikloheksilamin, cikloheksanmetilamin, benzilamin, feniletilamin, benzhidrilamin i *O*-benzilhidroksilamin hidroklorid) daje reaktivne spojeve 1-karbamoilbenzotriazole **7** (aktivne uree). Reakcija se provodi uz ekvimolarni omjer TEA koji veže klorovodik oslobođen u reakciji. Dvostruko više TEA potrebno je u reakciji dobivanja 1-(*N*-benziloksikarbamoil)benzotriazola (**7g**) gdje se drugi mol TEA troši za oslobađanje *O*-benzilhidroksilamina iz oblika soli. Svi produkti su dobiveni prema već opisanom postupku (Butula *et al*, 2000; Butula *et al*, 1977). Nastali 1--karbamoilbenzotriazoli **7** (aktivne uree) su reaktivni spojevi koji već u blagim uvjetima s aminima i srodnim spojevima reagiraju dajući uree, semikarbazide i karbazide, a s alkoholima karbamate (Butula *et al*, 2007).



	7a	**7b**	**7c**	**7d**	**7e**	**7f**	**7g**
R							

Shema 23. Sinteza 1-karbamoilbenzotriazola **7a-g**

Pripravljeni su sljedeći 1-karbamoilbenzotriazoli:

1-(*N*-ciklopentilkarbamoil)benzotriazol (**7a**),

1-(*N*-cikloheksilkarbamoil)benzotriazol (**7b**),

1-(*N*-cikloheksanmetilkarbamoil)benzotriazol (**7c**),

1-(*N*-benzilkarbamoil)benzotriazol (**7d**),

1-(*N*-feniletilkarbamoil)benzotriazol (**7e**),

1-(*N*-benzhidrilkarbamoil)benzotriazol (**7f**),

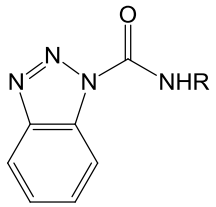
1-(*N*-benziloksikarbamoil)benzotriazol (**7g**).

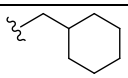
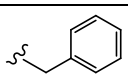
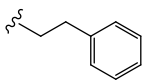
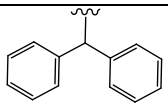
Od sintetiziranih spojeva novi spojevi su derivati **7a**,**c**,**d**,**f**. Sinteze svih derivata i njihova čistoća praćeni su tankoslojnom kromatografijom, a strukture su potvrđene IR, ^{1}H i ^{13}C NMR spektroskopijom te elementarnom analizom.

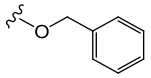
IR spektri 1-karbamoilbenzotriazola pokazuju karakteristične vrpce kod 3246–3369 (NH), 1707–1739 (C=O, amid I) i 1517–1603 (C=O, amid II) cm^{-1}. ^{1}H NMR spektri ispitivanih spojeva otkrivaju da se NH nalazi u području 8,98–10,00 ppm. Analizom ^{13}C NMR spektara utvrđeno je da se karbonil nalazi u području 148,61–149,67 ppm. Kemijski pomaci protona u ^{1}H, te atoma ugljika u ^{13}C NMR spektrima u skladu su s predloženim strukturama novih spojeva.

Kako u prijašnjim publikacijama nedostaju NMR spektroskopski podaci, u Tablicama 7–9 dan je pregled analitičkih i spektroskopskih podataka za sve spojeve **7**.

Tablica 7. Analitički podaci za 1-karbamoilbenzotriazole **7a-g**

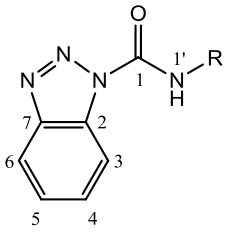


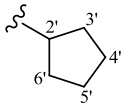
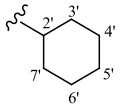
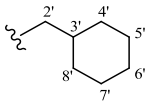
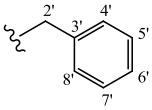
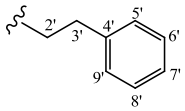
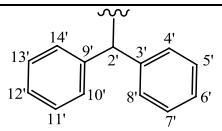
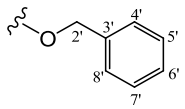
Spoj	**R**	t_t **(°C)**	**Mol. formula (M_r)**	**IR (KBr) ν_{max} (cm^{-1})**
7a		67–68	$C_{12}H_{14}N_4O$ (230,27)	3255, 2949, 2866, 1736, 1523, 1487, 1448, 1371, 1287, 1233, 1185, 1130, 1073, 1000, 944, 925, 815, 747, 652
7b		140[a]	$C_{13}H_{16}N_4O$ (244,29)	3327, 2938, 2856, 1733, 1517, 1488, 1447, 1360, 1285, 1226, 1151, 1090, 1059, 1002, 968, 928, 835, 791, 755, 612
7c		66–68	$C_{14}H_{18}N_4O$ (258,32)	3360, 2925, 2852, 1739, 1527, 1488, 1448, 1364, 1286, 1232, 1152, 1072, 1011, 954, 786, 771, 753, 593
7d		110	$C_{14}H_{12}N_4O$ (252,27)	3362, 1733, 1526, 1486, 1447, 1358, 1288, 1256, 1231, 1130, 1076, 1002, 793, 752, 696, 609, 580, 562
7e		114–115 115–117[b]	$C_{15}H_{14}N_4O$ (266,30)	3369, 3024, 2949, 1736, 1530, 1497, 1447, 1363, 1288, 1231, 1128, 1097, 1034, 1002, 920, 840, 749, 699, 588, 563
7f		120–122	$C_{20}H_{16}N_4O$ (328,37)	3288, 3028, 2931, 1707, 1520, 1449, 1375, 1293, 1231, 1069, 1031, 931, 864, 752, 695, 644, 607, 535

7g		113–114[c]	$C_{14}H_{12}N_4O_2$ (268,27)	3246, 3133, 3030, 2947, 2884, 1728, 1603, 1484, 1367, 1289, 1231, 1132, 1093, 1003, 924, 869, 751, 699, 563 (Butula *et al*, 2000)

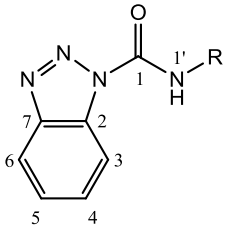
[a]Butula *et al*, 1977; [b]Butula *et al*, 1978; [c]Butula *et al*, 2000

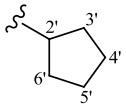
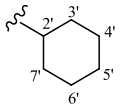
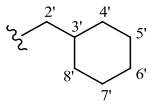
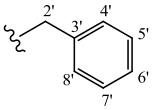
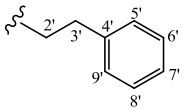
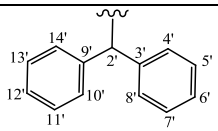
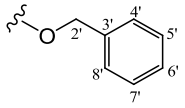
Tablica 8. ^{1}H NMR za 1-karbamoilbenzotriazole **7a-g**



Spoj	**R**	**^{1}H NMR (DMSO-d_6, δ ppm)**
7a		9,09 (d, 1H, 1', *J* = 6,62 Hz), 8,19 (d, 2H, arom., *J* = 8,27 Hz), 7,71, 7,54 (2t, 2H, *J* = 7,72 Hz, *J* = 6,62 Hz), 4,29–4,22 (m, 1H, 2'), 1,97–1,56 (m, 8H, 3'–6')
7b		8,98 (d, 1H, 1', *J* = 8,18 Hz), 8,19 (d, 2H, arom., *J* = 9,20 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,67 Hz), 3,81–3,73 (m, 1H, 2'), 1,93–1,11 (m, 10H, 3'–7')
7c		9,18 (t, 1H, 1', *J* = 5,62 Hz), 8,20 (d, 2H, arom., *J* = 9,20 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,67 Hz), 3,23 (t, 2H, 2', *J* = 6,14 Hz), 1,77–1,58, 1,27–0,93 (2m, 11H, 3'–8')
7d		9,78 (t, 1H, 1', *J* = 5,81 Hz), 8,20 (d, 2H, arom., *J* = 8,72 Hz), 7,72, 7,55 (2t, 2H, arom., *J* = 7,89 Hz), 7,45–7,25 (m, arom., 5H), 4,59 (d, 2H, 2', *J* = 6,23 Hz)
7e		9,27 (t, 1H, 1', *J* = 6,06 Hz), 8,21 (d, 2H, arom., *J* = 8,48 Hz), 7,71, 7,54 (2t, 2H, arom., *J* = 7,27 Hz), 7,34–7,19 (m, arom., 5H), 3,63 (q, 2H, 2', *J* = 6,67 Hz), 2,98 (t, 2H, 3', *J* = 7,88 Hz)
7f		10,00 (d, 1H, 1', *J* = 8,57 Hz), 8,21, 8,17 (2d, 2H, arom., *J* = 8,04 Hz), 7,74–7,28 (m, 12H, arom.), 6,40 (d, 1H, 2', *J* = 9,11 Hz)
7g		9,95 (s, 1H, 1'), 8,27, 8,09 (2d, 2H, *J* = 8,20 Hz, 8,50 Hz), 7,67, (t, 2H, *J* = 7,7 Hz), 7,48–7,34 (m, arom., 5H), 5,14 (s, 2H, 2') (Butula *et al*, 2000)

Tablica 9. ^{13}C NMR za 1-karbamoilbenzotriazole **7a-g**

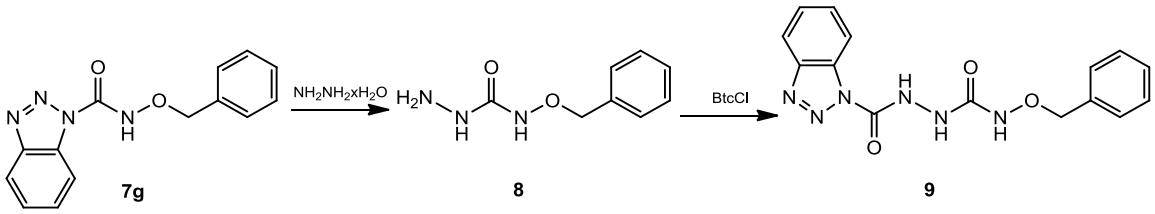


Spoj	R	^{13}C NMR (DMSO-d_6, δ ppm)
7a		148,98 (1), 145,87, 131,85 (2, 7), 130,24, 125,90, 120,16, 114,11 (3–6), 52,55 (2'), 32,18, 24,01 (3'–6')
7b		148,61 (1), 145,91, 131,89 (2, 7), 130,24, 125,94, 120,17, 114,05 (3–6), 50,34 (2'), 32,41, 25,35 (3', 4', 6', 7'), 25,46 (5')
7c		149,54 (1), 145,94, 131,83 (2, 7), 130,28, 125,90, 120,18, 114,05 (3–6), 40,60 (2'), 37,77 (3'), 30,76, 25,78 (4', 5', 7', 8'), 25,86 (6')
7d		149,67 (1), 145,96, 131,84 (2, 7), 139,10 (3'), 130,39, 125,98, 120,24, 114,02 (3–6), 128,84, 127,88 (4', 5', 7', 8'), 127,54 (6'), 43,94 (2')
7e		149,37 (1), 145,92, 131,78 (2, 7), 139,42 (4'), 130,34, 125,94, 120,20, 114,01 (3–6), 129,15, 128,85 (5', 6', 8', 9'), 126,69 (7'), 41,96, 35,36 (2', 3')
7f		149,16 (1), 145,93, 131,95 (2, 7), 141,62 (3', 9'), 130,42, 126,08, 120,27, 113,96 (3–6), 128,87, 128,23 (4', 5', 7', 8', 10', 11', 13', 14'), 127,82 (6', 12'), 58,37 (2')
7g		148,68 (1), 145,72, 131,54 (2, 7), 134,46 (3'), 130,23, 125,66, 120,07, 113,49 (3–6), 129,28, 128,63 (4', 5', 7', 8'), 126,97 (6'), 79,24 (2') (Butula *et al*, 2000)

3.1.3 Sinteza 4-benziloksisemikarbazida (8) i 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazida (9)

1-(*N*-Benziloksikarbamoil)benzotriazol **7g** korišten je za pripravu 4-benziloksisemikarbazida (**8**). BtH je u reakciji nukleofilne supstitucije zamijenjen hidrazinom. Reakcija je izvođena uz mali suvišak hidrazin hidrata da bi spriječili kondenzaciju nastalog semikarbazida sa spojem **7g**. Iz istog razloga je otopina spoja **7g** polako dokapavana u hidrazin hidrat, a ne obrnuto. U reakciji nije uočen nastanak mogućeg kondenziranog derivata.

Dobiveni semikarbazid je zatim u reakciji s kloridom **1** dao 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazid (**9**). Spoj **9** se u kasnijim reakcijama pokazao kao vrlo prikladan reagens za uvođenje acilhidroksisemikarbazidne skupine u molekule. Nije pokazao sklonost intramolekulskoj ciklizaciji.

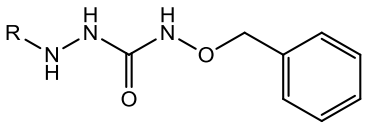


Shema 24. Sinteza 4-benziloksisemikarbazida **8** i 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazida **9**

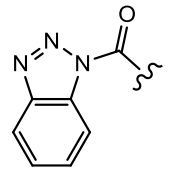
Sinteze novih spojeva i njihova čistoća praćeni su tankoslojnom kromatografijom, a strukture su potvrđene IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom.

IR spektri spojeva **8** i **9** pokazuju karakteristične vrpce kod 3164–3482 (NH). Vrpce karakteristične za karbonilne skupine se razlikuju, kod spoja **8** je 1655 (amid I) i 1517 (amid II) dok se kod spoja **9** osim tih vrpca 1674 (amid I) i 1556 (amid II) javlja još jedna vrpca karbonila na 1738 cm^{-1}. ^{1}H NMR spektri ispitivanih spojeva otkrivaju da se NH vezan na karbonile nalazi u području 7,84–10,97 ppm, a u spoju **8** se još jedan NH javlja na 4,01 ppm. Analizom ^{13}C NMR spektara utvrđeno je da se karbonil u položaju 3 nalazi u području 159,37–161,67 ppm, a karbonil u položaju 1' (spoj **9**) na 149,86 ppm. Kemijski pomaci protona u ^{1}H, te atoma ugljika u ^{13}C NMR spektrima u skladu su s predloženim strukturama novih spojeva. Analitički i spektroskopski podaci za nove spojeve dani su u Tablicama 10 i 11.

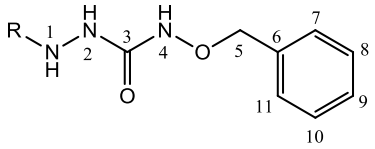
Tablica 10. Analitički i IR podaci za spojeve **8** i **9**

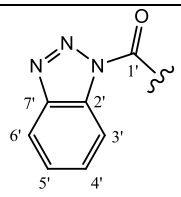


Spoj	**R**	t_t **(°C)**	**Mol. formula (M_r)**	**IR (KBr) ν_{max} (cm^{-1})**
8	H	90–92	$C_8H_{11}N_3O_2$ (181,19)	3332, 3287, 3217, 3061, 3030, 2962, 2925, 2875, 1655, 1637, 1517, 1456, 1366, 1325, 1228, 1210, 1192, 1096, 1070, 980, 915, 797,

				751, 702, 680, 607
9		90–93 (raspad)	$C_{15}H_{14}N_6O_3$ (326,31)	3482, 3376, 3340, 3164, 3005, 2924, 2854, 1738, 1674, 1556, 1487, 1449, 1387, 1287, 1235, 1066, 1033, 905, 754, 744, 704, 680, 652, 609, 580, 539

Tablica 11. ^{1}H i ^{13}C NMR podaci za spojeve **8** i **9**

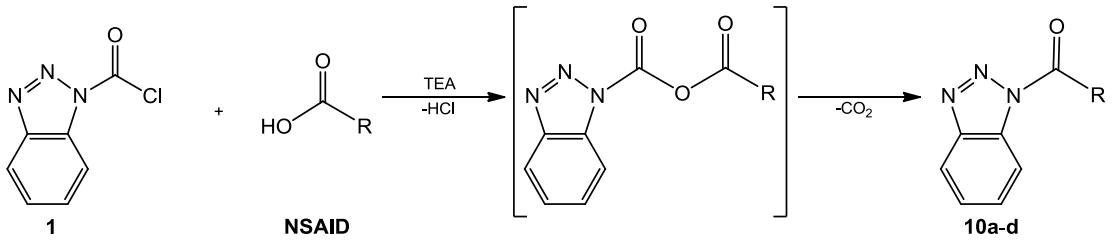


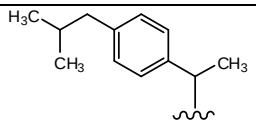
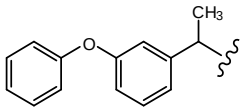
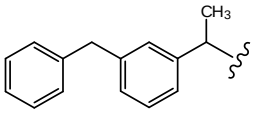
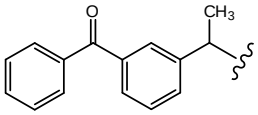
Spoj	**R**	**^{1}H i ^{13}C NMR (DMSO-d_6, δ ppm)**
8	H	9,18 (s, 1H, 4), 7,84 (s, 1H, 2), 7,42–7,29 (m, 5H, arom.), 4,71 (s, 2H, 5), 4,01 (s, 2H, 1) 161,67 (3), 137,18 (6), 129,08, 128,63 (7, 8, 10, 11), 128,37 (9), 77,70 (5)
9		10,97, 9,87, 9,31 (3s, 3H, 1, 2, 4), 8,26–8,18 (2d, 2H, arom.), 7,77, 7,59 (2t, 2H, arom.), 7,49–7,35 (m, 5H, arom., 7–11), 4,85 (s, 2H, 5) 159,37 (3), 149,86 (1'), 145,66, 131,92 (2', 7'), 136,74 (6), 130,78, 126,31, 120,42, 113,79 (3'–6'), 129,20, 128,68 (7, 8, 10, 11), 128,54 (9), 78,11 (5)

3.1.4. Sinteza derivata NSAID 10-14

3.1.4.1. *Sinteza NSAID benzotriazolida **10a-d***

NSAID benzotriazolidi **10a-d** pravljeni su reakcijom klorida **1** s odgovarajućim NSAID (ibuprofen, fenoprofen, ketoprofen i njegov reducirani derivat) u omjeru 1:1, u prisutnosti TEA (Rajić *et al*, 2010; Zorc *et al*, 1994; Zorc *et al*, 1993). Reakcijom prvo nastaje mješoviti anhidrid koji dekarboksilacijom daje benzotriazolid **10** (Shema 25). U benzotriazolidima je karboksilna skupina dovoljno aktivirana da s aminima i alkoholima daje amide, odnosno estere u blagim uvjetima, uz odcjepljenje BtH što je korišteno u sintezi svih derivata NSAID pripravljenih u okviru ove disertacije (Butula *et al*, 2007).



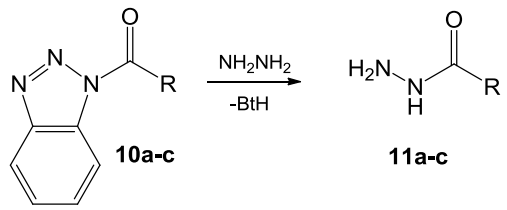
	10a	10b	10c	10d
R				

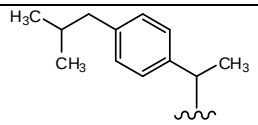
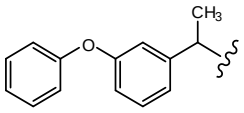
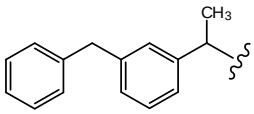
Shema 25. Sinteza NSAID benzotriazolida **10a-d**.

Sinteza i čistoća spojeva **10a-d** praćeni su tankoslojnom kromatografijom, a njihove strukture potvrđene su IR, ^{1}H i ^{13}C NMR spektroskopijom. Tališta ranije pripravljenih spojeva **10a-d** i njihovi spektroskopski podaci u skladu su s literaturnim podacima (Rajić *et al*, 2010; Zorc *et al*, 1994; Zorc *et al*, 1993).

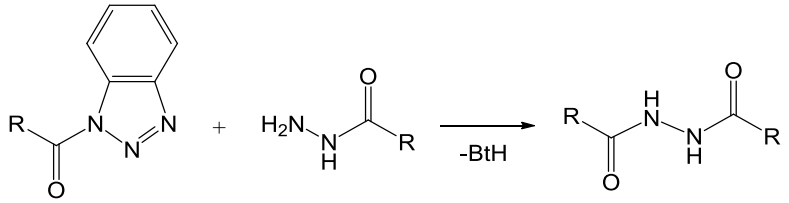
3.1.4.2. *Sinteza NSAID hidrazida **11a-c***

BtH je u spojevima **10a-c** u reakciji nukleofilne supstitucije zamijenjen hidrazinom (Shema 26). Reakcije su izvođene uz suvišak hidrazin hidrata da bi spriječili kondenzaciju nastalog produkta hidrazida s neizreagiranim benzotriazolidom **10** pri čemu može nastati diacil hidrazin (Shema 27).



	11a	11b	11c
R			

Shema 26. Sinteza NSAID hidrazida **11a-c**



Shema 27. Kondenzacija NSAID benzotriazolida i hidrazida u diacil hidrazine

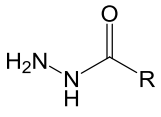
Pokušaj sinteze ketoprofenskog hidrazida nije uspio zbog reakcije keto skupine s hidrazin hidratom (Kürti *et al*, 2005) tako da je on isključen iz daljnih sinteza. U zamjenu je uzet njegov reducirani derivat (keto skupina reducirana do metilenske).

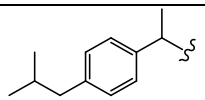
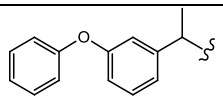
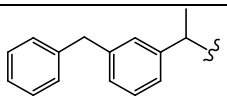
Priređeni su sljedeći NSAID hidrazidi:
2-(4-izobutilfenil)propanhidrazid (**11a**),
2-(3-fenoksifenil)propanhidrazid (**11b**),
2-(3-benzilfenil)propanhidrazid (**11c**).

Od sintetiziranih spojeva novi spojevi su **11b**,**c**, a svi su pripravljeni novom, do sada neobjavljenom metodom. Sinteza spojeva **11a-c** i njihova čistoća praćeni su tankoslojnom kromatografijom, a strukture potvrđene IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom. Tališta i spektroskopski podaci ranije pripravljenog derivata podudaraju se s literaturnim podacima (Sharma *et al*, 2003).

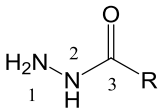
IR spektri ispitivanih spojeva pokazuju karakteristične vrpce kod 3023–3289 (NH) i 1637–1688 (C=O, amid I) i 1534–1582 (C=O, amid II) cm^{-1}. ^{1}H NMR spektri ispitivanih spojeva otkrivaju da se NH vezan na karbonil nalazi u području 9,12–9,14 ppm, a NH_2 se javlja na 4,15–4,25 ppm. Analizom ^{13}C NMR spektara utvrđeno je da se karbonil nalazi u području 172,87–173,37 ppm. Kemijski pomaci protona u ^{1}H, te atoma ugljika u ^{13}C NMR spektrima u skladu su s predloženim strukturama novih spojeva. Analitički i spektroskopski podaci prikazani su u Tablicama 12 i 13.

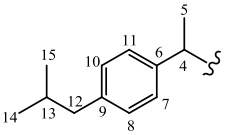
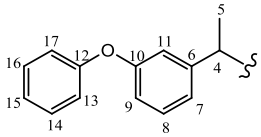
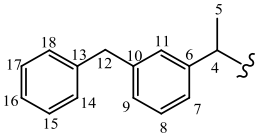
Tablica 12. Analitički i IR podaci za NSAID hidrazide **11a-c**



Spoj	R^1	t_t (°C)	Molekulska formula (M_r)	IR (KBr/film) ν_{max} (cm^{-1})
11a		80	$C_{13}H_{20}N_2O$ (220,31)	3276, 3180, 3052, 2954, 2919, 1688, 1641, 1535, 1510, 1452, 1416, 1358, 1257, 1237, 1124, 1074, 1007, 965, 848, 800, 786, 731, 690, 521
11b		ulje	$C_{15}H_{16}N_2O_2$ (256,30)	3285, 3040, 2978, 1659, 1631, 1582, 1487, 1445, 1244, 1210, 1164, 963, 928, 757, 692
11c		80–82	$C_{16}H_{18}N_2O$ (254,33)	3289, 3191, 3168, 3063, 3023, 2975, 2919, 1637, 1601, 1534, 1486, 1453, 1382, 1261, 1008, 976, 786, 759, 722, 696, 685, 599

Tablica 13. ^{1}H i ^{13}C NMR podaci za NSAID hidrazide **11a-c**



Spoj	R	^{1}H i ^{13}C NMR (DMSO-d_6, δ ppm)
11a		9,12 (s, 1H, 2), 7,12 (dd, 4H, arom., *J* = 8,32 Hz, 38,27 Hz), 4,15 (s, 2H, 1), 3,46 (q, 1H, 4, *J* = 6,96 Hz), 2,38 (d, 2H, 12, *J* = 6,96 Hz), 1,82–1,73 (m, 1H, 13), 1,30 (d, 3H, 5, *J* = 6,96 Hz), 0,83 (d, 6H, 14,15, *J* = 6,65 Hz) 173,37 (3), 139,78, 139,70 (6, 9), 129,15, 127,44 (7, 8, 10, 11), 44,70 (12), 43,37 (4), 30,09 (13), 22,63 (14, 15), 18,82 (5)
11b		9,19 (s, 1H, 2), 7,43–6,82 (m, 9H, arom.), 4,25 (s, 2H, 1), 3,53 (q, 1H, 4, *J* = 6,99 Hz), 1,32 (d, 3H, 5, *J* = 7,14 Hz) 172,87 (3), 157,01, 156,92, 144,73 (6, 10, 12), 130,50, 119,04 (13, 14, 16, 17), 130,15, 123,88, 122,86, 118,06, 117,02 (7–9, 11, 15), 43,58 (4), 18,76 (5)
11c		9,14 (s, 1H, 2), 7,31–7,03 (m, 9H, arom.), 4,17 (s, 2H, 1), 3,90 (s, 2H, 12), 3,48 (q, 1H, 4, *J* = 6,91 Hz), 1,33 (d, 3H, 5, *J* = 7,27 Hz) 173,19 (3), 142,66, 141,65, 141,47 (6, 10, 13), 129,14, 128,88 (13, 14, 16, 17), 128,69, 128,15, 127,38, 126,42, 125,44 (7–9, 11, 15), 43,72 (4), 41,64 (12), 18,88 (5)

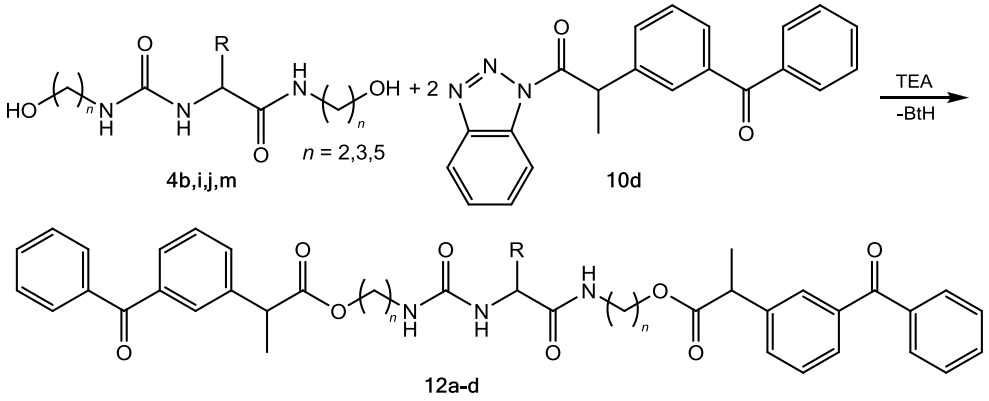
3.1.4.3. *Sinteza estera ketoprofena i ureidoamida **12a-d***

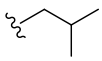
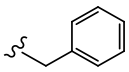
U sklopu ranijih istraživanja na Zavodu za farmaceutsku kemiju razvijena je metoda dobivanja estera iz NSAID benzotriazolida. Sintetizirani su metilni i etilni esteri te esterski polimer-lijek konjugati (Zorc *et al*, 1994; Zorc *et al*, 1993). Ta metoda korištena je i u sintezi estera **12a-d** (Shema 28) uz male modifikacije reakcijskih uvjeta čime je znatno skraćeno trajanje reakcije. Reakcije su provođene bez otapala na temperaturi taljenja smjese reaktanata uz suvišak TEA. Usporedba reakcijskih uvjeta dana je u tablici (Tablica 14).

Tablica 14. Usporedba različitih reakcijskih uvjeta u sintezi estera NSAID

	Otapalo	**NSAID-Bt/ TEA**	**Temp./°C**	**Trajanje reakcije/h**	**Iskorištenje/ %**
Metoda I (Zorc *et al*, 1993)	Metanol/etanol (ujedno i reagens)	1:4	Refluks	3	82–100
Metoda II (nova)	–	1:2	100–125[a]	0,25	65–82

[a] temperatura taljenja reakcijske smjese



	12a	**12b**	**12c**	**12d**
R	CH_3			
n	3	5	2	2

Shema 28. Sinteza estera ketoprofena **12a-d**

Priređeni su sljedeći esteri:

2-(3-benzilfenil)-*N*-ciklopentilpropanamid (1**2a**),

2-(3-benzilfenil)-*N*-cikloheksilpropanamid (**12b**),

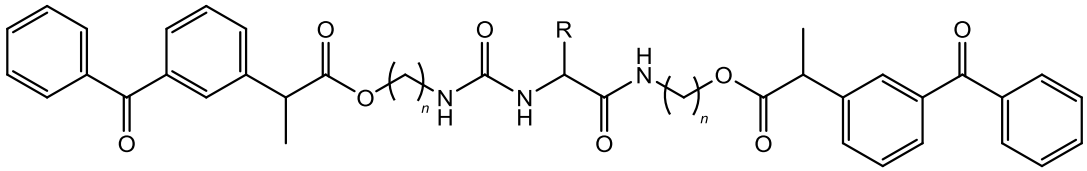
2-(3-benzilfenil)-*N*-(cikloheksanmetil)propanamid (**12c**),

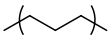
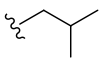
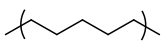
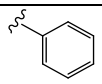
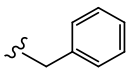
N-benzil-2-(3-benzilfenil)propanamid (**12d**).

Svi sintetizirani esteri su novi spojevi. Tijek sinteze i čistoća spojeva praćeni su tankoslojnom kromatografijom, a njihove strukture potvrđene su IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom.

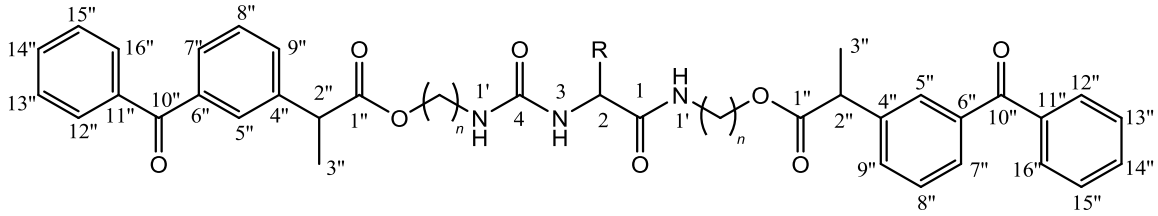
U IR spektrima prisutne su karakteristične vrpce za NH u području 3063–3302 i za karbonilne skupine u području 1731–1735 cm^{-1} (ester), 1633–1663 cm^{-1} (amid I) i 1556–1569 cm^{-1} (amid II). U ^{1}H NMR spektrima spojeva karbamidni NH se javljaju u području 5,92–6,36 ppm (1') i 5,87–6,84 ppm (3), a amidni NH u području 7,87–8,47 ppm (1'). U spojevima 12 nalazi se 4 vrste karbonilnih skupina: keto skupina na položaju 10" je u području 196,01–196,01 ppm, esterska na položaju 1" 173,85–174,00 ppm, amidna na položaju 1 171,40–173,60 ppm i karbamidna na položaju 4 157,44–157,95 ppm. Analitički i spektroskopski podaci dani su u Tablicama 15–17.

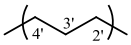
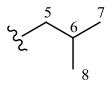
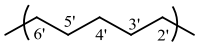
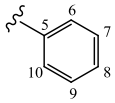
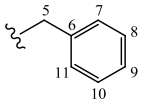
Tablica 15. Analitički i IR podaci za estere **12a-d**



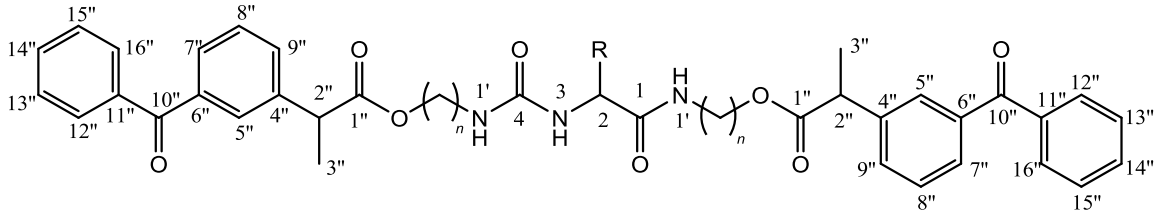
Spoj	**R**		**Mol. formula (M_r)**	**IR (NaCl/film) ν_{max} (cm^{-1})**
12a	CH_3		$C_{42}H_{45}N_3O_8$ (719,82)	3302, 3065, 2927, 2869, 1731, 1659, 1635, 1569, 1556, 1449, 1380, 1318, 1284, 1250, 1206, 1173, 1079, 960, 719, 644
12b			$C_{49}H_{59}N_3O_8$ (818,01)	3291, 3064, 2932, 2869, 1732, 1659, 1633, 1566, 1556, 1448, 1378, 1318, 1283, 1206, 1176, 1077, 954, 721, 706, 643
12c			$C_{45}H_{43}N_3O_8$ (753,84)	3296, 3064, 2980, 1735, 1633, 1564, 1449, 1374, 1284, 1172, 1077, 698, 643
12d			$C_{46}H_{45}N_3O_8$ (767,86)	3302, 3063, 2979, 2938, 1732, 1663, 1646, 1569, 1554, 1451, 1380, 1284, 1205, 1177, 1078, 956, 823, 722, 644

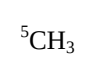
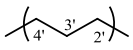
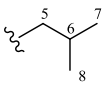
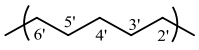
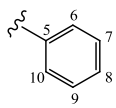
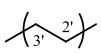
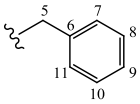
Tablica 16. ^{1}H NMR podaci za estere **12a-d**



Spoj	R		^{1}H NMR (DMSO-d_6, δ ppm, J/Hz)
12a			7,91 (t, 1H, 1'(amid), J = 5,62 Hz), 7,74–7,50 (m, 18H, arom.), 6,08 (t, 1H, 1'(urea), J = 5,62 Hz), 6,01 (d, 1H, 3, J = 7,79 Hz), 4,10–3,90 (m, 7H, 2, 4', 2"), 3,08–2,93 (m, 4H, 2'), 1,70–1,59 (m, 4H, 3'), 1,43 (d, 6H, 3", J = 7,07 Hz), 1,11 (d, 3H, 5, J = 6,89 Hz)
12b			7,87 (t, 1H, 1'(amid), J = 5,50 Hz), 7,73–7,50 (m, 18H, arom.), 5,92 (t, 1H, 1'(urea), J = 5,50 Hz), 5,87 (d, 1H, 3, J = 8,84 Hz), 4,13–3,89 (m, 7H, 2, 6', 2"), 3,02–2,86 (m, 4H, 2'), 1,54–1,13 (m, 21H, 5, 6, 3'–5', 3"), 0,84 (2d, 6H, 7, 8, J = 5,85 Hz)
12c			8,47 (t, 1H, 1'(amid), J = 4,34 Hz), 7,75–7,49 (m, 18H, arom.), 7,37–7,22 (m, 5H, arom.), 6,84 (d, 1H, 3, J = 8,31 Hz), 6,36 (t, 1H, 1'(urea), J = 5,54 Hz), 5,32 (d, 1H, 2, J = 8,01 Hz), 4,10–3,78 (m, 6H, 3', 2"), 3,32–3,21 (m, 4H, 2'), 1,43, 1,37 (2d, 6H, 3", J = 7,32 Hz)
12d			8,14 (t, 1H, 1'(amid), J = 5,25 Hz), 7,74–7,48 (m, 18H, arom.), 7,22–7,09 (m, 5H, arom.), 6,22–6,18 (d+t, 2H, 3, 1' (urea)), 4,36 (q, 1H, 2, J = 6,44 Hz), 4,07–3,88 (m, 6H, 3', 2"), 3,31–3,14 (m, 4H, 5, 2'), 2,88–2,66 (m, 2H, 2'), 1,44–1,41 (m, 6H, 3")

Tablica 17. ^{13}C NMR podaci za estere **12a-d**



Spoj	R		^{13}C NMR (DMSO-d_6, δ ppm, J/Hz)
12a			196,07 (10"), 174,00, 173,95 (1"), 173,60 (1), 157,72 (4), 141,58, 137,65, 137,41 (4", 8", 11"), 133,20, 132,22, 129,30, 128,95, 128,90, (5"–7", 9", 14"), 130,05, 129,04 (12", 13", 15", 16"), 62,82, 62,73 (4'), 49,01 (2), 44,71 (2"), 36,23, 35,57 (2'), 29,61, 28,70 (3'), 20,19 (5), 18,91 (3")
12b			196,04 (10"), 173,95 (1"), 173,30 (1), 157,95 (4), 141,62, 137,63, 137,41 (4", 8", 11"), 133,21, 132,20, 129,36, 128,92, (5"–7", 9", 14"), 130,03, 129,04 (12", 13", 15", 16"), 64,75 (6'), 51,89 (2), 44,75 (2"), 43,00 (5), 39,43, 38,56 (2'), 29,95, 28,95, 28,18, 28,09, 23,05, 23,00 (3'–5'), 24,73, 23,38, 22,54 (6–8), 18,82 (3")
12c			196,04 (10"), 173,93, 173,85 (1"), 171,40 (1), 157,44 (4), 141,47, 141,39, 140,72, 140,70, 140,66, 137,62, 137,39 (5, 4", 8", 11",), 133,20, 132,31, 132,27, 129,28, 128,94, (5"–7", 9", 14"), 130,08, 129,04 (12", 13", 15", 16"), 127,64, 127,01 (6, 7, 9, 10), 127,68 (8), 64,59, 63,32 (3'), 56,97 (2), 44,75, 44,68 (2"), 38,59, 38,03 (2'), 18,99, 18,89 (3")
12d			196,04, 196,01 (10"), 173,93 (1"), 172,59 (1), 157,73 (4), 141,48, 141,46, 138,17, 138,14, 137,62, 137,61, 137,39 (6, 4", 8", 11"), 133,20, 132,30, 129,29, 128,96 (5"–7", 9", 14"), 130,07, 129,03 (12", 13", 15", 16"), 129,67, 128,39 (7, 8, 10, 11), 126,59 (9), 64,54 63,54, 63,40 (3'), 54,69 (2), 44,74 (2"), 39,34 (5), 38,55, 37,90 (2'), 19,00 (3")

3.1.4.4. *Sinteza 4-supstituiranih 1-acilsemikarbazida NSAID **13a-y***

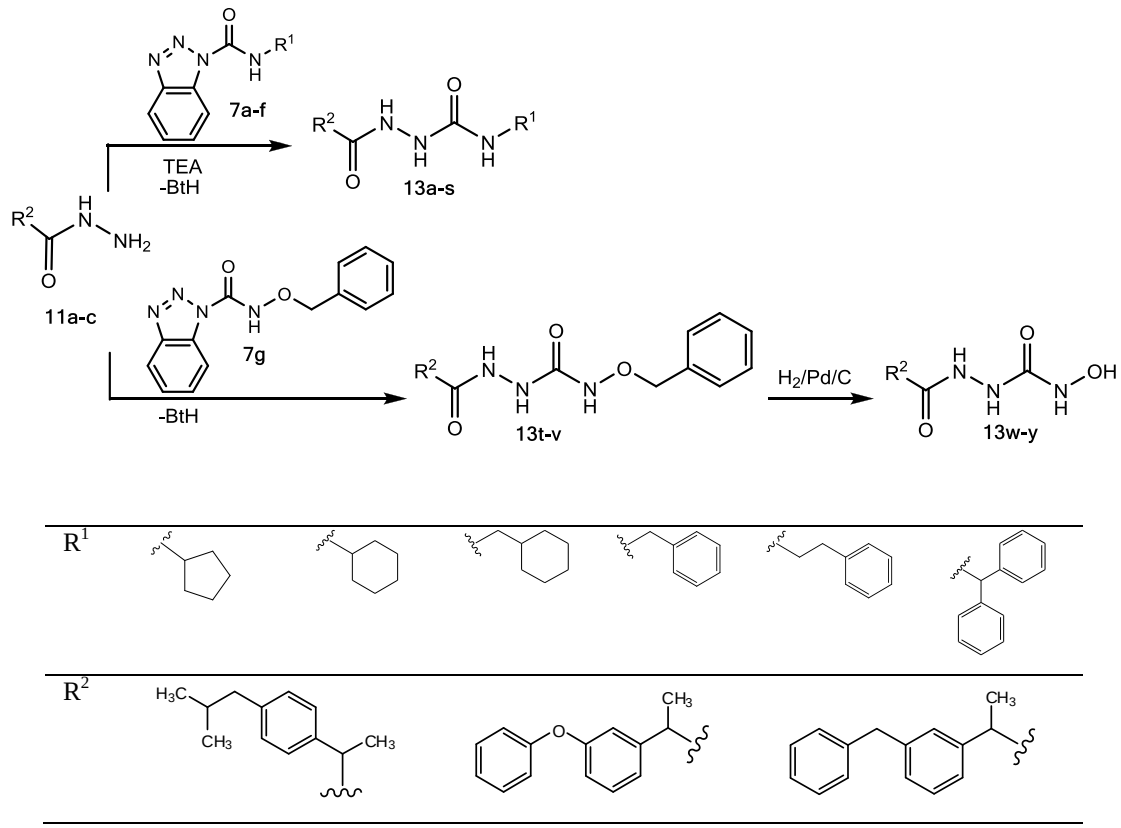
Semikarbazidni derivati NSAID **13** (Shema 29) pripravljeni su iz NSAID hidrazida **11** i odgovarajućeg 1-karbamoilbenzotriazola **7**. Ovisno o supstituentu na položaju 4 semikarbazidne okosnice upotrebljeni su različiti reakcijski uvjeti. Tako su dobiveni cikloaklil/aril (**13a-s**), benziloksi (**13t-v**) i hidroksi (**13w-y**) derivati. 1-Acil-4-hidroksisemikarbazidi **13w-y** dobiveni su hidrogenolizom benzilne skupine benziloksisemikarbazida **13t-v**.

Sinteza 4-cikloalkil/aril derivata **13a-s** izvođene su uz suvišak TEA, na minimalnoj temperaturi taljenja smjese reaktanata (75–120 °C) i bez otapala. Takvi uvjeti omogućili su brz završetak reakcije (15 minuta) i pojednostavljenu obradu reakcijske smjese (ekstrakcija u lužnatom radi uklanjanja oslobođenog benzotriazola). U reakcijama nije uočena ciklizacija niti raspad bilo reaktanata bilo produkata.

Suprotno tome, derivati **13t-v** s benziloksi skupinom u položaju 4 pripravljeni su na nižoj temperature (55 °C), u otapalu (dioksan) i bez TEA. Razlog tome je mogućnost ciklizacije koja se često javlja kod spojeva s *O*-benzilnom skupinom, a koja je u blažim reakcijskim uvjetima uspješno izbjegnuta. Zato su te reakcije trajale puno dulje (6–8 sati). Iskorištenja su podjednaka za oba načina sinteze. Uvjeti obrade reakcija nakon njihovog završetka se također razlikuju, ovisno o topljivosti nastalog produkta u etil-acetatu koji je korišten kao organski sloj u ekstrakciji. Ukoliko produkt nije bio topljiv, ekstrakcija je zamijenjena obradom u smjesi acetona i zakiseljene vode čime je uspješno uklonjen BtH oslobođen u reakciji. U Tablici 18 dan je pregled reakcijskih uvjeta i pripadajućih iskorištenja za sve semikarbazidne derivate.

Tablica 18. Reakcijski uvjeti za spojeve **13**

Spoj 13	**Množinski omjer reaktanata (7:11:TEA)**	**Otapalo**	**Temp. (°C)**	**Trajanje reakcije (h)**	**Obrada**	**Iskorištenje (%)**
a,b,e-j,l-n,r,s	1:1:3	–	75–120	0,25	ekstrakcija u lužnatom	47–85
c,d,k,o,p	1:1:3	–	75–120	0,25	smjesa acetona i zakiseljene vode	63–94
t-v	1:1:0	dioksan	55	6–8	ekstrakcija u lužnatom	50–91
w-y		metanol	s.t.	1,5	–	75–96



Shema 29. Sinteza 1-acilsemikarbazida NSAID **13a-y**

Pripravljeni su sljedeći 1-acilsemikarbazidi:

4-ciklopentil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13a**),

4-cikloheksil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13b**),

4-cikloheksanmetil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13c**),

4-benzil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13d**),

4-feniletil-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13e**),

4-benzhidril-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13f**),

4-ciklopentil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13g**),

4-cikloheksil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13h**),

4-cikloheksanmetil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13i**),

4-cenzil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13j**),

4-feniletil-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13k**),

4-benzhidril-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13l**),

4-ciklopentil-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13m**),

4-cikloheksil-2-[2-(3-benzilfenil)propanoil]semikarbazid (**13n**),

4-cikloheksanmetil-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13o**),

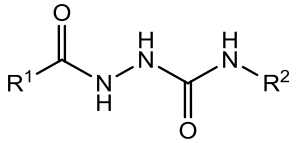
4-benzil-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13p**),
4-feniletil-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13r**),
4-benzhidril-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13s**),
4-benziloksi-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13t**),
4-benziloksi-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13u**),
4-benziloksi-1-[2-(3-benzilfenil)propanoil]semikarbazid (**13v**),
4-hidroksi-1-[2-(4-izobutilfenil)propanoil]semikarbazid (**13w**),
4-hidroksi-1-[2-(3-fenoksifenil)propanoil]semikarbazid (**13x**),
4-hidroksi-1-(2-(3-benzilfenil)propanoil]semikarbazid (**13y**).

Tijek sinteze i čistoća spojeva praćeni su tankoslojnom kromatografijom, a njihove strukture potvrđene su IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom.

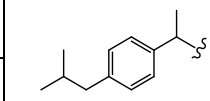
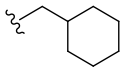
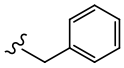
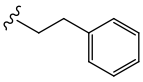
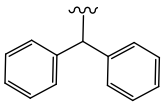
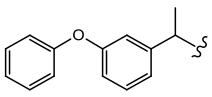
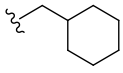
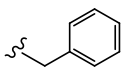
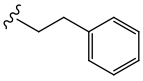
IR spektri semikarbazida **13** pokazuju karakteristične vrpce kod 3025–3472 (NH i OH), 1601–1699 (amid I) i 1506–1590 (amid II) cm^{-1}. U ^{1}H NMR spektrima derivata **13a-s** signali NH na položajima 1 i 2 javljaju se u području 9,63–9,78 i 7,59–7,89 ppm, a na položaju 2' ima širi raspon od 5,71–6,99 ppm (pomaci u niže polje posljedica su blizine aromatskih prstenova koji ju otkrivaju) u derivatima **d-f**, **j-l** i **p-s**. U derivatima **13t-z** zbog elektron odvlačećeg utjecaja atoma kisika, signali NH su pomaknuti u niže polje osim najdaljeg NH na položaju 2 koji se kao i kod derivata **a-s** nalazi u području 9,72–9,74 ppm, dok su preostala dva NH u derivatima **t-v** nalaze u području 9,46–9,47 i 8,69–8,71 ppm. Derivatima **w-z** NH na položajima 1 i 2' te OH nalaze se u području 8,41–8,73 ppm. Karbonilna skupina na položaju 3 u ^{13}C NMR spektrima nalazi se u području 172,59–175,27 ppm, a na položaju 1' u području 157,02–160,82 ppm.

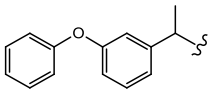
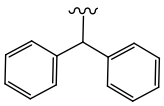
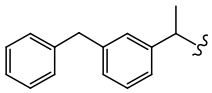
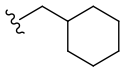
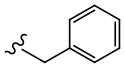
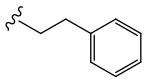
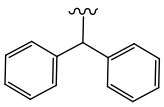
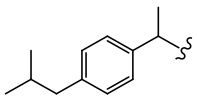
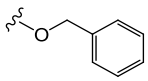
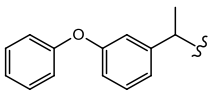
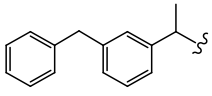
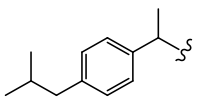
Svi spojevi serije **13** su novi. Njihovi analitički i spektroskopski podaci dani su u Tablicama 19–21.

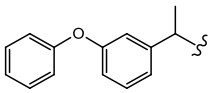
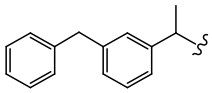
Tablica 19. Analitički i IR podaci za 1-acilsemikarbazide NSAID **13a-y**



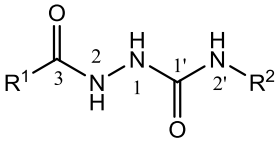
Spoj	**R^{1}**	**R^{2}**	**t_t (°C)**	**Mol. formula (M_r)**	**IR (KBr) ν_{max} (cm^{-1})**

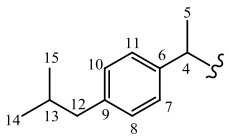
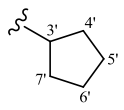
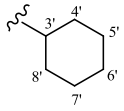
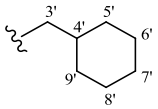
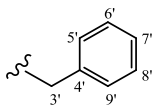
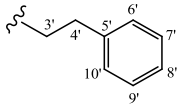
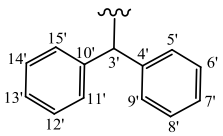
13a			123–128	$C_{19}H_{29}N_3O_2$ (331,45)	3355, 3254, 2956, 2871, 1684, 1650, 1557, 1513, 1465, 1315, 1243, 1151, 1079, 1008, 937, 850, 786, 655
13b			165–166	$C_{20}H_{31}N_3O_2$ (345,48)	3307, 3253, 3019, 2986, 2951, 2930, 2852, 1708, 1683, 1641, 1562, 1531, 1506, 1453, 1322, 1253, 1226, 1153, 1079, 1056, 1023, 999, 936, 892, 850, 709, 634, 614
13c			172–174	$C_{21}H_{33}N_3O_2$ (359,51)	3339, 3236, 3122, 2952, 2926, 2849, 1699, 1615, 1565, 1476, 1448, 1368, 1263, 1247, 1175, 1084, 1019, 854, 780, 651, 618
13d			173–175	$C_{21}H_{27}N_3O_2$ (353,46)	3316, 3222, 3123, 3028, 2954, 1654, 1611, 1566, 1469, 1454, 1364, 1236, 1171, 1077, 851, 749, 699, 668
13e			93–97	$C_{22}H_{29}N_3O_2$ (367,48)	3566, 3434, 3312, 3026, 2953, 2926, 2869, 1648, 1587, 1557, 1514, 1497, 1455, 1374, 1253, 1168, 1074, 1054, 999, 934, 853, 796, 746, 700, 547
13f			151–154	$C_{27}H_{31}N_3O_2$ (429,55)	3343, 3202, 3028, 2953, 2869, 1647, 1616, 1538, 1495, 1456, 1366, 1268, 1230, 1168, 1077, 1052, 1028, 1004, 942, 848, 746, 700, 669, 631
13g			133–135	$C_{21}H_{25}N_3O_3$ (367,44)	3414, 3281, 3237, 3101, 3025, 2961, 2910, 2869, 1696, 1627, 1638, 1582, 1556, 1532, 1489, 1455, 1443, 1311, 1244, 1213, 1162, 1146,1131, 1073, 942, 913, 880, 774, 760, 691, 618
13h			144–146	$C_{22}H_{27}N_3O_3$ (381,47)	3343, 3215, 2933, 2853, 1695, 1618, 1580, 1555, 1490, 1472, 1448, 1314, 1273, 1235, 1213, 1166, 1074, 1062, 1025, 944, 887, 748, 689, 640
13i			104–105	$C_{23}H_{29}N_3O_3$ (395,49)	3339, 3230, 3115, 3026, 2924, 2850, 1701, 1619, 1582, 1568, 1490, 1447, 1373, 1246, 1212, 1166, 1146, 1074, 1024, 1012, 962, 933, 888, 749, 691, 668
13j			154–156	$C_{23}H_{23}N_3O_3$ (389,45)	3323, 3266, 3209, 3109, 3028, 1654,1621, 1584, 1565, 1490, 1453, 1369, 1315, 1278, 1249, 1235, 1162, 1143, 1071, 935,876, 751, 694, 644
13k			116–118	$C_{24}H_{25}N_3O_3$ (403,47)	3367, 3236, 3026, 2942, 1682, 1642, 1583, 1560, 1542, 1488, 1454, 1379, 1310, 1247, 1211, 1162, 1075, 1012, 944, 916, 866, 796, 747, 698, 637

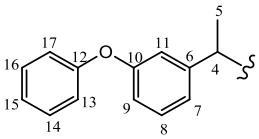
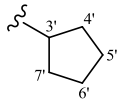
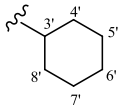
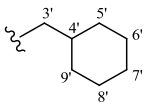
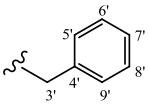
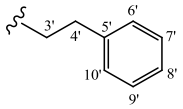
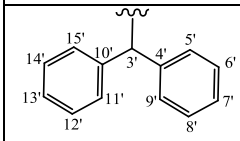
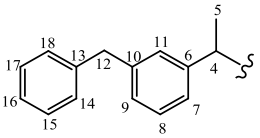
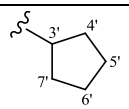
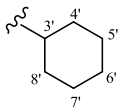
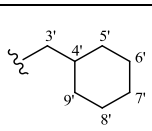
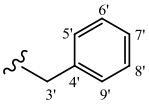
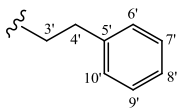
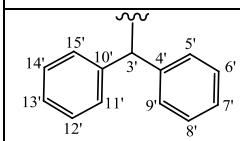
13l			126–131	$C_{29}H_{27}N_3O_3$ (465,54)	3350, 3236, 3031, 2973, 1655, 1613, 1587, 1560, 1488, 1448, 1247, 1210, 1164, 1070, 1052, 1030, 949, 912, 753, 698, 668, 634, 614
13m			133–134	$C_{22}H_{27}N_3O_2$ (365,47)	3279, 3027, 2962, 2872, 1710, 1646, 1555, 1494, 1452, 1372, 1242, 1191, 1152, 1077, 1010, 941, 728, 698, 638
13n			143–144	$C_{23}H_{29}N_3O_2$ (379,50)	3327, 3216, 3027, 2932, 2853, 1658, 1617, 1589, 1477, 1452, 1255, 1232, 1182, 1074, 1065, 943, 891, 769, 751, 701, 639,618
13o			133–137	$C_{24}H_{31}N_3O_2$ (393,52)	3344, 3216, 3027, 2919, 2850, 1663, 1622, 1589, 1474, 1450, 1370, 1273, 1257, 1239, 1184, 1075, 1030, 963, 746, 723, 702, 652, 618
13p			150–154	$C_{24}H_{25}N_3O_2$ (387,47)	3331, 3261, 3211, 3028, 2986, 1707, 1660, 1620, 1563, 1494, 1453, 1431, 1371, 1246, 1174, 1075, 750, 723, 698, 642, 618
13r			118–120	$C_{25}H_{27}N_3O_2$ (401,50)	3365, 3301, 3239, 3027, 2939, 1686, 1649, 1601, 1555, 1495, 1455, 1379, 1269, 1242, 1154, 1073, 945, 751, 726, 699, 661
13s			166–170	$C_{30}H_{29}N_3O_2$ (463,57)	3346, 3231, 3028, 1981, 1662, 1611, 1590, 1560, 1494, 1472, 1453, 1372, 1258, 1236, 1078, 1030, 948, 751, 700, 668, 634, 613
13t			87 (raspad)	$C_{21}H_{27}N_3O_3$ (369,46)	3472, 3319, 3240, 3114, 3064, 3031, 2953, 2867, 1714, 1698, 1681, 1638, 1594, 1548, 1514, 1455, 1366, 1349, 1226, 1147, 1082, 994, 933, 910, 850, 802, 751, 698, 620, 592, 545
13u			ulje	$C_{23}H_{23}N_3O_4$ (405,45)	3237, 3063, 3034, 2979, 2934, 2877, 1668, 1583, 1488, 1455, 1371, 1312, 1244, 1211, 1163, 1073, 942, 914, 753, 695, 614
13v			104–105	$C_{24}H_{25}N_3O_3$ (403,47)	3306, 3235, 3029, 2925, 2880, 1691, 1654, 1599, 1520, 1493, 1454, 1382, 1366, 1356, 1273, 1238, 1152, 1074, 1032, 1007, 947, 788, 751, 727, 699, 668, 555, 532, 497
13w		-OH	123–125	$C_{14}H_{21}N_3O_3$ (279,33)	3330, 3235, 3027, 2954, 2924, 2869, 1704, 1652, 1556, 1514, 1465, 1383, 1320, 1262, 1141, 1078, 1002, 935, 850, 766, 727, 653, 531

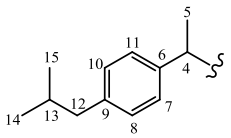
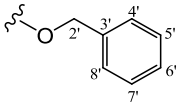
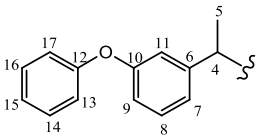
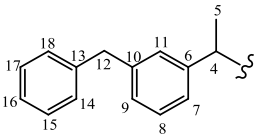
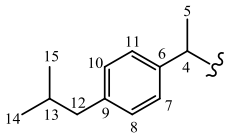
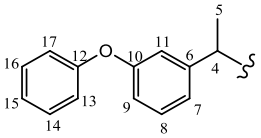
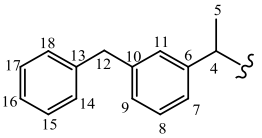
13x		-OH	150 (raspad)	$C_{16}H_{17}N_3O_4$ (315,32)	3298, 3214, 3026, 2983, 2941, 1685, 1643, 1582, 1567, 1489, 1446, 1362, 1313, 1268, 1232, 1212, 1161, 1140, 1080, 1023, 969, 939, 876, 788, 756, 693, 649, 596, 558, 523
13y			123–125	$C_{17}H_{19}N_3O_3$ (313,35)	3288, 3060, 3028, 2979, 2938, 1687, 1644, 1562, 1516, 1494, 1483, 1451, 1380, 1263, 1159, 1144, 1100, 1077, 1061, 1027, 938, 782, 721, 700, 596, 533

Tablica 20. ^{1}H NMR podaci za 1-acilsemikarbazide NSAID **13a**-**y**

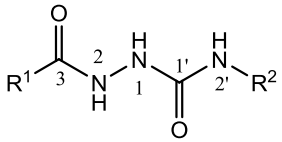


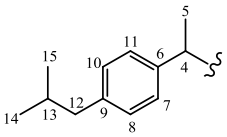
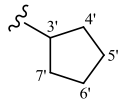
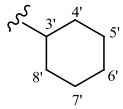
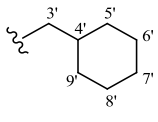
Spoj	R^1	R^2	^{1}H NMR (DMSO-d_6, δ ppm)
13a			9,63, 7,59, (2s, 2H, 1, 2), 7,15 (dd, 4H, arom, *J* = 7,55 Hz, 87,09 Hz), 6,50 (bs, 1H, 2'), 3,84–3,80 (m, 1H, 3'), 3,59 (q, 1H, 4, *J* = 7,02 Hz), 2,40 (d, 2H, 12, *J* = 7,17 Hz), 1,84–1,20 (m, 9H, 13, 4'–7'), 1,32 (d, 3H, 5, *J* = 7,02 Hz), 0,85 (d, 6H, 14,15, *J* = 6,58 Hz)
13b			9,67, 7,64, (2d, 2H, 1, 2, *J* = 1,77 Hz), 7,15 (dd, 4H, arom., *J* = 7,76 Hz, 39,23 Hz), 5,71 (d, 1H, 2', *J* = 6,43 Hz), 3,59 (q, 1H, 4, *J* = 6,87 Hz), 3,39–3,28 (m, 2H, 3'), 2,40 (d, 2H, 12, *J* = 7,10 Hz), 1,84–1,75 (m, 1H, 13), 1,69–0,95 (m, 10H, 4'–8'), 1,32 (d, 3H, 5, *J* = 6,87 Hz), 0,85 (d, 6H, 14, 15, *J* = 6,65 Hz)
13c			9,65 7,66 (2s, 2H, 1, 2), 7,15 (dd, 4H, arom., *J* = 7,74 Hz, 40,61 Hz), 5,99 (t, 1H, 2', *J* = 4,78 Hz), 3,59 (q, 1H, 4, *J* = 6,86 Hz), 2,82 (t, 2H, 3', *J* = 6,22 Hz), 2,40 (d, 2H, 12, *J* = 7,02 Hz), 1,84–1,75 (m, 1H, 13), 1,66–0,78 (m, 11H, 4'–9'), 1,33 (d, 3H, 5, *J* = 6,53 Hz), 0,85 (d, 6H, 14, 15, *J* = 5,87 Hz)
13d			9,70, 7,85 (2s, 2H, 1, 2), 7,32–7,05 (m, 9H, arom.), 6,70 (t, 1H, 2', *J* = 6,01 Hz), 4,22 (d, 2H, 3', *J* = 6,01 Hz), 3,59 (q, 1H, 4, *J* = 6,94 Hz), 2,39 (d, 2H, 12, *J* = 6,94 Hz), 1,83–1,74 (m, 1H, 13), 1,34 (d, 3H, 5, *J* = 6,94 Hz), 0,84 (d, 6H, 14, 15, *J* = 6,48 Hz)
13e			9,66, 7,79 (2s, 2H, 1, 2), 7,38–7,00 (m, 9H, arom.), 6,15 (t, 1H, 2', *J* = 5,52 Hz), 3,59 (q, 1H, 4, *J* = 6,81 Hz), 3,22 (q, 2H, 3', *J* = 6,49 Hz), 2,67 (t, 2H, 4', *J* = 7,14 Hz), 2,40 (d, 2H, 12, *J* = 6,81 Hz), 1,85–1,76 (m, 1H, 13), 1,34 (d, 3H, 5, *J* = 7,14 Hz), 0,86 (d, 6H, 14, 15, *J* = 6,49 Hz)
13f			9,76, 7,84 (2s, 2H, 1, 2), 7,34–7,05 (m, 14H, arom.), 6,95 (bs, 1H, 2'), 5,91 (d, 1H, 3', *J* = 8,38 Hz), 3,58 (q, 1H, 4, *J* = 6,85 Hz), 2,39 (d, 2H, 12, *J* = 7,11 Hz), 1,83–1,74 (m, 1H, 13), 1,33 (d, 3H, 5, *J* = 6,85 Hz), 0,84 (d, 6H, 14, 15, *J* = 6,60 Hz)

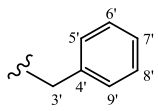
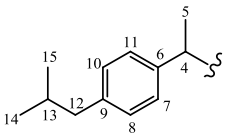
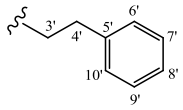
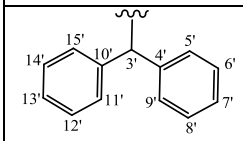
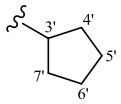
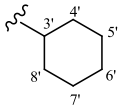
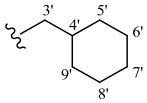
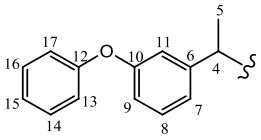
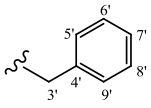
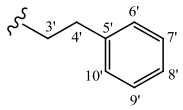
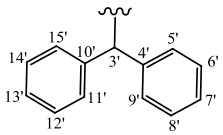
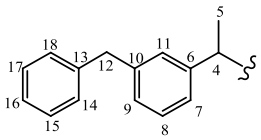
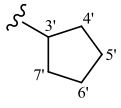
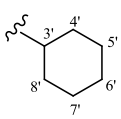
13g			9,69, 7,63 (2s, 2H, 1, 2), 7,42–6,82 (m, 9H, arom.), 5,94 (d, 1H, 2', *J* = 6,73 Hz), 3,89–3,78 (m, 2H, 3'), 3,63 (q, 1H, 4, *J* = 6,73 Hz), 1,79–1,22 (m, 8H, 4'–7'), 1,32 (d, 3H, 5, *J* = 6,73 Hz)
13h			9,70, 7,66 (2s, 2H, 1, 2), 7,42–6,83 (m, 9H, arom.), 5,84 (d, 1H, 2', *J* = 7,00 Hz), 3,63 (q, 1H, 4, *J* = 6,73 Hz), 3,39–3,33 (m, 2H, 3'), 1,71–1,00 (m, 10H, 4'–8'), 1,33 (d, 3H, 5, *J* = 7,00 Hz)
13i			9,70, 7,70 (2s, 2H, 1, 2), 7,43–6,84 (m, 9H, arom.), 6,09 (t, 1H, 2', *J* = 5,20 Hz), 3,63 (q, 1H, 4, *J* = 6,82 Hz), 2,84 (t, 2H, 3', *J* = 6,17 Hz), 1,62–0,79 (m, 11H, 4'–9'), 1,34 (d, 3H, 5, *J* = 6,82 Hz)
13j			9,74, 7,89 (2s, 2H, 1, 2), 7,41–6,82 (m, 14H, arom.), 6,76 (t, 1H, 2', *J* = 5,58 Hz), 4,22 (d, 2H, 3', *J* = 5,58 Hz), 3,63 (q, 1H, 4, *J* = 6,50 Hz), 1,34 (d, 3H, 5, *J* = 6,81 Hz)
13k			9,70, 7,82 (2s, 2H, 1, 2), 7,42–6,84 (m, 14H, arom.), 6,21 (t, 1H, 2', *J* = 5,36 Hz), 3,63 (q, 1H, 4, *J* = 6,80 Hz), 3,23 (q, 2H, 3', *J* = 6,59 Hz), 2,68 (t, 2H, 4', *J* = 7,21 Hz), 1,34 (d, 3H, 5, *J* = 7,00 Hz)
13l			9,78, 7,85 (2s, 2H, 1, 2), 7,38–6,98 (m, 19H, arom.), 6,83 (d, 1H, 2', *J* = 7,36 Hz), 5,91 (d, 1H, 3', *J* = 8,83 Hz), 3,64 (q, 1H, 4, *J* = 6,31 Hz), 1,33 (d, 3H, 5, *J* = 6,75 Hz)
13m			9,67, 7,60 (2s, 2H, 1, 2), 7,31–7,05 (m, 9H, arom.), 5,90 (d, 1H, 2', *J* = 6,27 Hz), 3,91 (s, 2H, 12), 3,82 (m, 1H, 3'), 3,59 (q, 1H, 4, *J* = 6,86 Hz), 1,76–1,22 (m, 8H, 4'–7'), 1,32 (d, 3H, 5, *J* = 7,05 Hz)
13n			9,68, 7,64 (2s, 2H, 1, 2), 7,30–7,05 (m, 9H, arom.), 5,80 (d, 1H, 2', *J* = 7,29 Hz), 3,91 (s, 2H, 12), 3,59 (q, 1H, 4, *J* = 6,97 Hz), 3,38–3,29 (m, 1H, 3'), 1,70–0,97 (m, 10H, 4'–8'), 1,32 (d, 3H, 5, *J* = 6,97 Hz)
3o			9,67, 7,66 (2s, 2H, 1, 2), 7,31–7,05 (m, 9H, arom.), 6,04 (t, 1H, 2', *J* = 5,23 Hz), 3,91 (s, 2H, 12), 3,59 (q, 1H, 4, *J* = 6,82 Hz), 2,82 (t, 2H, 3', *J* = 6,14 Hz), 1,66–0,78 (m, 11H, 4'–9'), 1,32 (d, 3H, 5, *J* = 6,82 Hz)
13p			9,72, 7,86 (2s, 2H, 1, 2), 7,32–7,04 (m, 14H, arom.), 6,73 (t, 1H, 2', *J* = 6,19 Hz), 4,22 (d, 2H, 3', *J* = 5,77 Hz), 3,89 (s, 2H, 12), 3,60 (q, 1H, 4, *J* = 7,01 Hz), 1,34 (d, 3H, 5, *J* = 7,01 Hz)
13r			9,66, 7,78 (2s, 2H, 1, 2), 7,29–7,04 (m, 14H, arom.), 6,17 (t, 1H, 2', *J* = 5,61 Hz), 3,90 (s, 2H, 12), 3,58 (q, 1H, 4, *J* = 6,83 Hz), 3,21 (q, 2H, 3', *J* = 6,58 Hz), 2,65 (t, 2H, 4', *J* = 7,31 Hz), 1,32 (d, 3H, 5, *J* = 7,07 Hz)
13s			9,78, 7,84 (2s, 2H, 1, 2), 7,33–7,00 (m, 19H, arom.), 6,99 (d, 1H, 2', *J* = 8,85 Hz), 5,91 (d, 1H, 3', *J* =8,38 Hz), 3,89 (s, 2H, 12), 3,60 (q, 1H, 4, *J* = 6,99 Hz), 1,33 (d, 3H, 5, *J* = 6,99 Hz)

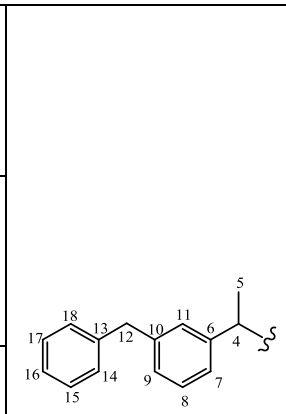
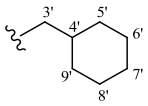
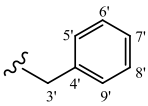
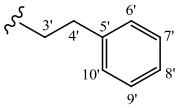
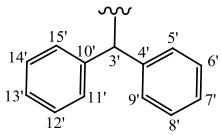
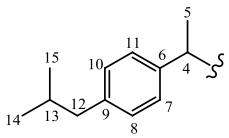
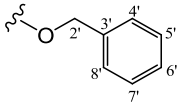
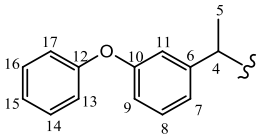
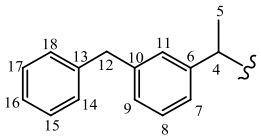
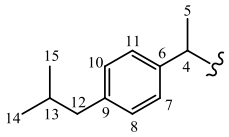
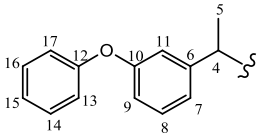
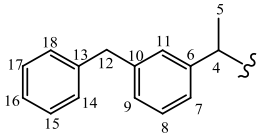
13t			9,73, 9,47, 8,71 (3s, 3H, 1, 2, 2'), 7,43–7,32 (m, 5H, arom.), 7,17 (dd, 4H, arom., *J* = 8,03 Hz, 45,43 Hz), 4,74 (s, 2H, 3'), 3,62 (q, 1H, 4, *J* = 6,81 Hz), 2,40 (d, 2H, 12, *J* = 6,99 Hz), 1,87–1,76 (m, 1H, 13), 1,34 (d, 3H, 5, *J* = 7,16 Hz), 0,85 (d, 6H, 14, 15, *J* = 6,46 Hz)
13u			9,74, 9,47, 8,70 (3s, 3H, 1, 2, 2'), 7,41–6,83 (m, arom., 14H), 4,75 (s, 2H, 3'), 3,66 (q, 1H, 4, *J* = 6,94 Hz), 1,35 (d, 3H, 5, *J* = 6,94 Hz)
13v			9,72, 9,46, 8,69 (3s, 3H, 1, 2, 2'), 7,37–7,02 (m, arom., 14H), 4,72 (s, 2H, 3'), 3,88 (s, 2H, 12), 3,59 (q, 1H, 4, *J* = 6,46 Hz), 1,31 (d, 3H, 5, *J* = 6,85 Hz)
13w		-OH$^{3'}$	9,65, 8,70, 8,64, 8,41 (4s, 4H, 1, 2, 2', 3'), 7,15 (dd, 4H, arom, *J* = 7,92 Hz, 46,47 Hz), 3,61 (q, 1H, 4, *J* = 6,89 Hz), 2,40 (d, 2H, 12, *J* = 7,11 Hz), 1,85–1,76 (m, 1H, 13), 1,33 (d, 3H, 5, *J* = 6,89 Hz), 0,85 (d, 6H, 14, 15, *J* = 6,45 Hz)
13x			9,68, 8,72, 8,65, 8,45 (4s, 4H, 1, 2, 2', 3'), 7,42–6,82 (m, arom., 9H), 3,65 (q, 1H, 4, *J* = 6,95 Hz), 1,33 (d, 3H, 5, *J* = 6,95 Hz)
13y			9,66, 8,73, 8,66, 8,43 (4s, 4H, 1, 2, 2', 3'), 7,32–7,05 (m, arom., 9H), 3,92 (s, 2H, 12), 3,62 (q, 1H, 4, *J* = 6,81 Hz), 1,34 (d, 3H, 5, *J* = 7,01 Hz)

Tablica 21. ^{13}C NMR podaci za 1-acilsemikarbazide NSAID **13a-y**



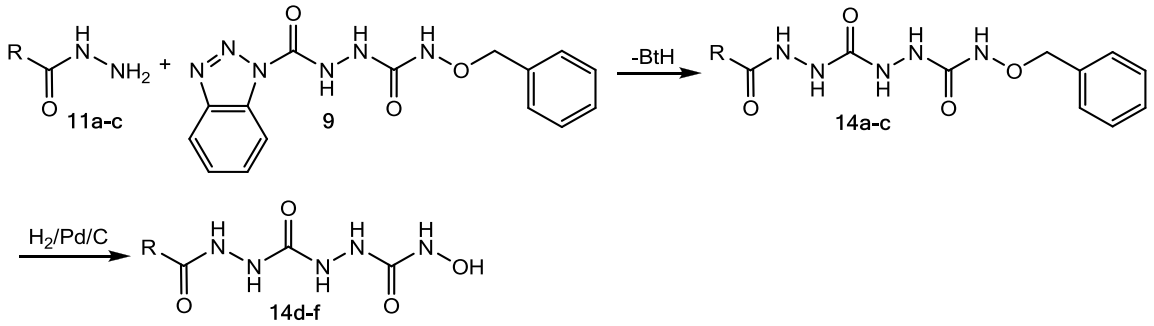
Spoj	R^1	R^2	^{13}C NMR (DMSO-d_6, δ ppm)
13a			175,27 (3), 159,64 (1'), 141,54, 141,08 (6, 9), 130,96, 129,18 (7, 8, 10, 11), 53,09 (3'), 46,42 (12), 44,76 (4), 34,86, 25,33 (4'–7'), 31,76 (13), 24,34 (14, 15), 20,37 (5)
13b			173,56 (3), 157,60 (1'), 139,84, 139,36 (6, 9), 129,27, 127,47 (7, 8, 10, 11), 48,28 (3'), 44,70 (12), 43,03 (4), 33,42, 33,38, 24,90 (4', 5', 7', 8'), 30,09 (13), 25,65 (6'), 22,64 (14, 15), 18,64 (5)
13c			173,66 (3), 158,47 (1'), 139,79, 139,35 (6, 9), 129,22, 127,50 (7, 8, 10, 11), 45,80 (3'), 44,71 (12), 43,06 (4), 38,37 (4'), 30,67, 30,65, 25,88 (5', 6', 8', 9'), 30,09 (13), 26,56 (7'), 22,64 (14, 15), 18,69 (5)

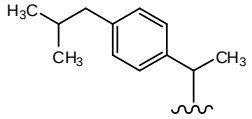
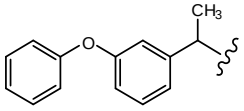
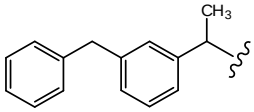
13d			173,83 (3), 158,59 (1'), 140,90 (4'), 139,78, 139,34 (6, 9), 129,21, 128,58, 127,55, 127,35, (7, 8, 10, 11, 5', 6', 8', 9'), 127,00 (7'), 44,70 (12), 43,11 (4), 43,07 (3'), 30,08 (13), 22,64 (14, 15), 18,81 (5)
13e			173,72 (3), 158,36 (1'), 139,99, 139,79, 139,33 (6, 9, 5'), 129,21, 129,10, 128,78, 127,54, (7, 8, 10, 11, 6', 7', 9', 10'), 126,48 (8'), 44,70 (12), 43,06 (4), 41,32 (3'), 36,38 (4'), 30,08 (13), 22,64 (14, 15), 18,78 (5)
13f			173,70 (3), 157,58 (1'), 143,51, 143,46 (4', 10'), 139,83, 139,26 (6, 9), 129,25, 128,81, 128,79, 127,49, 127,47, 127,44 (7, 8, 10, 11, 5', 6', 8', 9', 11', 12', 14', 15'), 127,35, 127,31 (7', 13'), 57,14 (3'), 44,71 (12), 43,05 (4), 30,06 (13), 22,65 (14, 15), 18,78 (5)
13g			173,08 (3), 157,89 (1'), 156,99, 156,94, 144,29 (6, 10, 12), 130,49, 119,01 (13, 14, 16, 17), 130,23, 123,87 , 122,93, 118,17, 117,12 (7–9, 11, 15), 51,40 (3'), 43,25 (4), 33,13, 24,91 (4'–7'), 18,66 (5)
13h			173,08 (3), 157,55 (1'), 156,99, 156,94, 144,29 (6, 10, 12), 130,49, 119,01 (13, 14, 15, 16), 130,24, 123,87 , 122,95, 118,16, 117,12 (7–9, 11, 15), 48,33 (3'), 43,24 (4), 33,41, 24,91 (4', 5', 7', 8'), 25,65 (6'), 18,66 (5)
13i			173,18 (3), 158,43 (1'), 157,03, 156,91, 144,29 (6, 10, 12), 130,19, 118,98 (13, 14, 16, 17), 130,21, 123,85 , 122,98, 118,23, 117,14 (7–9, 11, 15), 45,82 (3'), 43,29 (4), 38,38 (4'), 30,68, 25,88 (5', 6', 8', 9'), 26,56 (7'), 18,70 (5)
13j			173,35 (3), 158,54 (1'), 157,03, 156,90, 144,28 (6, 10, 12), 140,91 (4'), 130,49, 128,59, 127,34, 119,00 (13, 14, 16, 17, 5', 6', 8', 9'), 130,20, 127,00, 123,84, 123,02, 118,29, 117,14 (7–9, 11, 15, 7'), 43,36 (4), 43,08 (3'), 18,77 (5)
13k			173,23 (3), 158,31 (1'), 157,03, 156,90, 144,29 (6, 10, 12), 139,99 (5'), 130,49, 129,11, 128,79, 119,00, (13, 14, 16, 17, 6', 7', 9', 10'), 130,21, 126,48, 123,85, 123,02, 118,26, 117,14 (7–9, 11, 15, 8'), 43,29 (4), 41,32 (3'), 36,38 (4'), 18,78 (5)
13l			172,70 (3), 157,02 (1'), 156,50 156,47, 143,70 (6, 10, 12), 142,96, 143,03 (4', 10'), 129,97, 118,53 (13, 14, 16, 17), 129,73, 123,35, 122,43, 117,71, 116,65 (7–9, 11, 15), 128,37, 128,33, 126,97, 126,93 (5', 6', 8', 9', 11', 12', 14', 15'), 126,85, 126,81 (7',13'), 56,71 (3'), 42,80 (4), 18,24 (5)
13m			173,40 (3), 157,93 (1'), 142,22, 141,61, 141,55 (6, 10, 13), 129,14, 128,86 (14, 15, 17, 18), 128,76, 128,25, 127,48, 126,42, 125,45 (7–9, 11, 16), 51,38 (3'), 43,36 (4), 41,65 (12), 33,13, 23,62 (4'–7'), 18,73 (5)
13n			173,40 (3), 157,60 (1'), 142,22, 141,61, 141,55 (6, 10, 13), 129,14, 128,86 (14, 15, 17, 18), 128,77, 128,24, 127,48, 126,42, 125,45 (7–9, 11, 16), 48,31 (3'), 43,36 (4), 41,65 (12), 33,38, 24,89 (4', 5', 7', 8'), 25,66 (6'), 18,73 (5)

13o			173,48 (3), 158,47 (1'), 142,21, 141,62, 141,52 (6, 10, 13), 129,13, 128,86 (14, 15, 17, 18), 128,74, 128,24, 127,46, 126,41, 125,47 (7–9, 11, 16), 45,81 (3'), 43,38 (4), 41,65 (12), 38,36 (4'), 30,66, 25,88 (5', 6', 8', 9'), 26,55 (7'), 18,76 (5)
13p			173,67 (3), 158,59 (1'), 142,23, 141,63, 141,52 (6, 10, 13), 140,92 (4'), 129,15, 128,87, 128,59, 127,35 (14, 15, 17, 18, 5', 6', 8', 9'), 128,74, 128,28, 127,46, 127,01, 126,41, 125,55 (7–9, 11, 16, 7'), 43,46 (4), 43,08 (3'), 41,64 (12), 18,88 (5)
13r			173,55 (3), 158,37 (1'), 142,23, 141,63, 141,52 (6, 10, 13), 140,00 (5'), 129,15, 129,11, 128,87, 128,79 (14, 15, 17, 18, 6', 7', 9', 10'), 128,75, 128,28, 127,46, 126,48, 126,41, 125,54 (7–9, 11, 16, 8'), 43,41 (4), 41,65 (12), 41,33 (3'), 36,39 (4'), 18,87 (5)
13s			173,67 (3), 157,56 (1'), 143,52, 143,47 (4', 10'), 142,13, 141,59, 141,56, (6, 10, 13), 129,14, 128,86, 128,82, 128,80, 127,45 (14, 15, 17, 18, 5', 6', 8', '9', 11', 12', 14', 15'), 128,22, 127,34, 127,32, 126,40, 125,47 (7–9, 11, 16, 7', 13'), 57,16 (3'), 43,48 (4), 41,63 (12), 18,84 (5)
13t			173,55 (3), 159,52 (1'), 139,77, 139,25 (6, 9), 136,90 (4'), 129,17, 129,12, 128,61, 127,62 (7, 8, 10, 11, 5', 6', 8', 9'), 128,42 (7'), 77,90 (3'), 44,71 (12), 43,05 (4), 30,10 (13), 22,65 (14,15), 18,93 (5)
13u			172,59 (3), 158,95 (1'), 156,57, 156,38, 143,70 (6, 10, 12), 136,37 (4'), 129,98, 128,62, 128,11, 118,48 (13, 14, 16, 17, 5', 6', 8', 9'), 129,65, 127,93, 123,32, 122,59, 117,87, 116,65 (7–9, 11, 15, 7'), 77,42 (3'), 42,80 (4), 18,37 (5)
13v			173,40 (3), 159,52 (1'), 142,13, 141,65, 141,47 (6, 10, 13), 136,90 (4'), 129,16, 129,13, 128,87, 128,61 (14, 15, 17, 18, 5', 6', 8', 9'), 128,70, 128,43, 128,34, 127,44, 126,41, 125,61 (7–9, 11, 16, 7'), 77,90 (3'), 43,39 (4), 41,65 (12), 19,00 (5)
13w		-OH$^{3'}$	173,45 (3), 160,82 (1'), 139,71, 139,33 (6, 9), 129,14, 127,62 (7, 8, 10,11), 44,71 (12), 43,00 (4), 30,09 (13), 22,65 (14,15), 19,01 (5)
13x			172,98 (3), 160,78 (1'), 157,06, 156,84, 144,29 (6, 10, 12), 130,49, 118,98 (13, 14,16, 17), 130,13, 123,81, 123,10, 118,36, 117,11 (7–9, 11, 15), 43,24 (4), 18,95 (5)
13y			173,29 (3), 160,82 (1'), 142,22, 141,66, 141,45 (6, 10, 13), 129,16, 128,87 (14, 15, 17, 18), 128,68, 128,34, 127,38, 126,40, 125,60 (7–9, 11, 16), 43,34 (4), 41,65 (12), 19,08 (5)

3.1.4.5. *Sinteza 1-acil-5-benziloksi i 1-acil-5-hidroksihidroksikarbamoilkarbazida NSAID* ***14a-f***

Serija derivata NSAID bogata dušikom i kisikom pripravljena je aminolizom 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazida **9** s NSAID hidrazidima **11a-c**. Time su dobiveni benziloksi derivati **14a-c** koji su hidrogenolizom dali hidroksi derivate **14d-f** (Shema 30). Za razliku od reakcija **13a-s** u kojima je korišten TEA kao katalizator, u sintezi derivata **14a-c** TEA je izostavljen zbog moguće ciklizacije spoja **9**. Reakcije su također provođene u talini na minimalnoj temperaturi taljenja smjese reaktanata (90 °C) i bez otapala. U reakcijama nije uočena ciklizacija niti raspad bilo reaktanata bilo produkata.



	14a,d	14b,e	14c,f
R			

Shema 30. Sinteza 1-acil-5-benziloksi i 1-acil-5-hidroksikarbamoilkarbazida NSAID **14a-f**

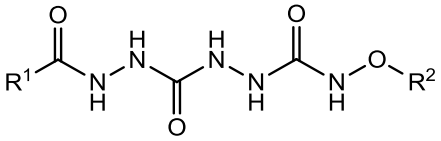
Pripravljeni su sljedeći 1-acilkarbamoilkarbazidi:
5-benziloksikarbamoil-1-[2-(4-izobutilfenil)propanoil]karbazid (**14a**),
5-benziloksikarbamoil-1-[2-(3-fenoksifenil)propanoil]karbazid (**14b**),
5-benziloksikarbamoil-1-[2-(3-benzilfenil)propanoil]karbazid (**14c**),
5-hidroksikarbamoil-1-[2-(4-izobutilfenil)propanoil]karbazid (**14d**),
5-hidroksikarbamoil-1-[2-(3-fenoksifenil)propanoil]karbazid (**14e**),
5-hidroksikarbamoil-1-[2-(3-benzilfenil)propanoil]karbazid (**14f**).

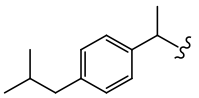
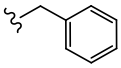
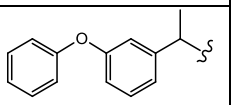
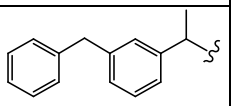
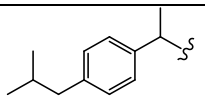
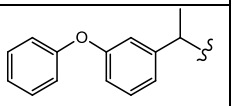
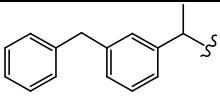
Tijek sinteze i čistoća spojeva praćeni su tankoslojnom kromatografijom, a njihove strukture potvrđene su IR, ^{1}H i ^{13}C NMR spektroskopijom i elementarnom analizom.

IR spektri derivata **14a-f** pokazuju karakteristične vrpce kod 3032–3266 (NH i OH), 1602–1677 (C=O, amid I) i 1513–1583 (C=O, amid II) cm^{-1}. U ^{1}H NMR spektrima NH i OH javljaju se u području 7,92–9,80 ppm. Karbonilna skupina na položaju 3 u ^{13}C NMR spektrima nalazi se u području 172,90–173,52 ppm, a na položaju 1' i 4' u području 158,30–161,24 ppm.

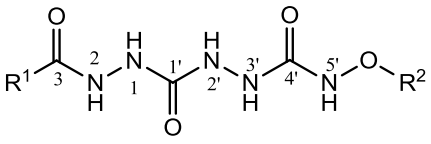
Svi spojevi serije **14** su novi. Njihovi analitički i spektroskopski podaci dani su u Tablicama 22–24.

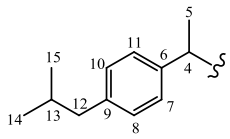
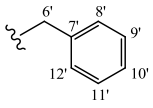
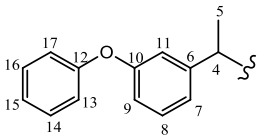
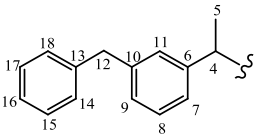
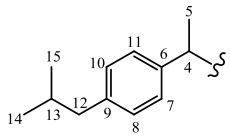
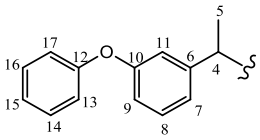
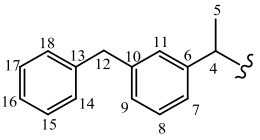
Tablica 22. Analitički i IR podaci za 1-acilkarbamoilkarbazide NSAID **14a-f**



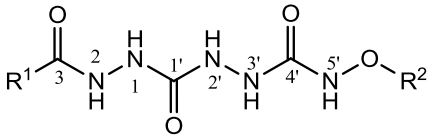
Spoj	R^1	R^2	t_t (°C)	Mol. formula (M_r)	IR (KBr) ν_{max} (cm^{-1})
14a			85–90	$C_{22}H_{29}N_5O_4$ (427,50)	3266, 3032, 2955, 2930, 2869, 1677, 1513, 1466, 1455, 1366, 1309, 1271, 1212, 1188, 1075, 1030, 1002, 934, 909, 849, 751, 699, 620, 547
14b			72–76	$C_{24}H_{25}N_5O_5$ (463,49)	3251, 3062, 3034, 2971, 2933, 2873, 1677, 1583, 1523, 1488, 1455, 1369, 1312, 1243, 1211, 1163, 1073, 1023, 1002, 942, 914, 816, 752, 694, 614
14c			73–77	$C_{25}H_{27}N_5O_4$ (461,51)	3248, 3060, 3028, 2880, 2836, 1675, 1602, 1519, 1494, 1453, 1369, 1310, 1270, 1210, 1075, 1030, 974, 941, 908, 751, 699, 620, 556
14d		H	145–148 (raspad)	$C_{15}H_{23}N_5O_4$ (337,37)	3261, 2955, 2928, 2870, 1670, 1514, 1466, 1383, 1367, 1281, 1206, 1077, 1002, 934, 850, 783
14e			100–103 (raspad)	$C_{17}H_{19}N_5O_5$ (373,36)	3263, 2978, 2925, 2855, 1670, 1583, 1540, 1488, 1456, 1445, 1376, 1313, 1244, 1210, 1163, 1074, 943, 917, 755, 692
14f			101–105 (raspad)	$C_{18}H_{21}N_5O_4$ (371,39)	3258, 3060, 3027, 2981, 2934, 1670, 1601, 1538, 1494,1453, 1375, 1321, 1195, 1075, 1030, 943, 784, 728, 699, 622, 557

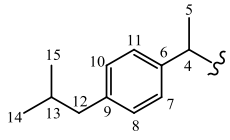
Tablica 23. ^{1}H NMR podaci za 1-acilkarbamoilkarbazide NSAID **14a-f**

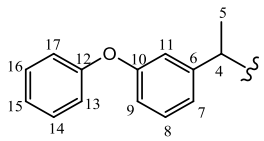
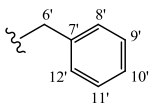
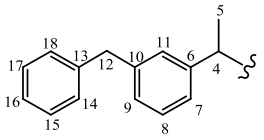
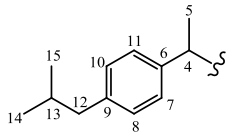
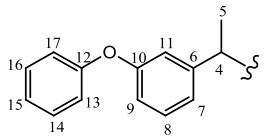
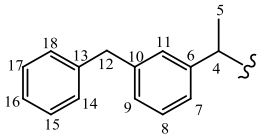


Spoj	R^1	R^2	^{1}H NMR (DMSO-d_6, δ ppm)
14a			9,75, 9,43, 8,54, 8,15, 8,03 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–7,30 (m, 5H, arom., 8'–12'), 7,16 (dd, 4H, arom., *J* = 7,99 Hz, 46,08 Hz), 4,77 (s, 2H, 6'), 3,62 (q, 1H, 4, *J* = 6,94 Hz), 2,41 (d, 2H, 12, *J* = 7,15 Hz), 1,85–1,76 (m, 1H, 13), 1,35 (d, 3H, 5, *J* = 6,94 Hz), 0,86 (d, 6H, 14, 15, *J* = 6,52 Hz)
14b			9,78, 9,43, 8,55, 8,18, 8,05 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–6,82 (m, 14H, arom.), 4,77 (s, 2H, 6'), 3,65 (q, 1H, 4, *J* = 6,97 Hz), 1,34 (d, 3H, 5, *J* = 7,12 Hz)
14c			9,76, 9,43, 8,55, 8,16, 8,03 (5s, 5H, 1, 2, 2', 3', 5'), 7,44–7,05 (m, 14H, arom.), 4,78 (s, 2H, 6'), 3,92 (s, 2H, 12), 3,62 (q, 1H, 4, *J* = 6,77 Hz), 1,34 (d, 3H, 5, *J* = 7,02 Hz)
14d		$H^{6'}$	9,73, 8,66, 8,57, 8,23, 8,08, 7,92 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,15 (dd, 4H, arom., *J* = 7,93 Hz, 95,99 Hz), 3,61 (q, 1H, 4, *J* = 7,01 Hz), 2,40 (d, 2H, 12, *J* = 7,28 Hz), 1,82–1,78 (m, 1H, 13), 1,33 (d, 3H, 5, *J* = 7,01 Hz), 0,85 (d, 6H, 14, 15, *J* = 6,47 Hz)
14e			9,80, 8,67, 8,29, 8,14, 7,96 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,42–6,81 (m, 9H, arom.), 3,64 (q, 1H, 4, *J* = 7,03 Hz), 1,34 (d, 3H, 5, *J* = 6,95 Hz)
14f			9,78, 8,67, 8,28, 8,11, 7,95 (5s, bs, 6H, 1, 2, 2', 3', 5', 6'), 7,31–7,04 (m, 9H, arom.), 3,91 (s, 2H, 12), 3,61 (q, 1H, 4, *J* = 6,88 Hz), 1,33 (d, 3H, 5, *J* = 7,14 Hz)

Tablica 24. ^{13}C NMR podaci za 1-acilkarbamoilkarbazide NSAID **14a-f**



Spoj	R^1	R^2	^{13}C NMR (DMSO-d_6, δ ppm)
14a			173,52 (3), 160,03, 158,37 (1', 4'), 139,75, 139,34, (6, 9), 136,97 (7'), 129,17, 129,14, 128,61, 127,59 (7, 8, 10, 11, 8', 9', 11', 12'), 128,41 (10'), 77,87 (6'), 44,71 (12), 43,00 (4), 30,09 (13), 22,65 (14, 15), 18,85 (5)

14b			173,03 (3), 160,03, 158,32 (1', 4'), 157,05, 156,87, 144,28 (6, 10, 12), 136,97 (7'), 130,50, 129,14, 128,61, 118,99 (13, 14, 16, 17, 8', 9', 11', 12'), 130,16, 128,41, 123,83, 123,06, 118,33,117,13 (7–9, 11, 15, 10'), 77,88 (6'), 43,25 (4), 18,79 (5)
14c			173,35 (3), 160,02, 158,36 (1', 4'), 142,21, 141,64, 141,48 (6, 10, 13), 136,96 (7'), 129,15, 128,87, 128,61 (14, 15, 17, 18, 8', 9', 11', 12'), 128,70, 128,41, 128,31, 127,42, 126,40, 125,58 (7–9, 11, 16, 10'), 77,88 (6'), 43,34 (4), 41,65 (12), 18,91 (5)
14d		$H^{6'}$	172,90 (3), 160,70, 157,83 (1', 4'), 139,24, 138,85 (6, 9), 128,6, 127,07 (7, 8, 10, 11), 44,22 (12), 42,49 (4), , 29,55 (13), 22,14 (14, 15), 18,35 (5)
14e			172,92 (3), 161,24, 158,30 (1', 4'), 157,03, 156,88, 144,28, (6, 10, 12), 130,50, 119,01 (13, 14, 16, 17), 130,16, 123,84, 123,03, 118,30, 117,11 (7–9, 11, 15), 43,22 (4), 18,78 (5)
14f			173,23 (3), 161,24, 158,34 (1', 4'), 142,22, 141,64, 141,48 (6, 10, 13), 129,16, 128,88 (14, 15, 17, 18), 128,71, 128,29, 127,42, 126,41, 125,56 (7–9, 11, 16), 43,32 (4), 41,65 (12), 18,90 (5)

3.2. BIOLOŠKA ISPITIVANJA

3.2.1. Antitumorska ispitivanja

Antitumorska ispitivanja provedena su na Institutu Rudjer Bošković i na Rega Institute for Medical Research, Katholieke Universiteit Leuven, Belgija prema ranije opisanoj metodi (Perković *et al*, 2010; Perković *et al*, 2008).

3.2.1.1. *Ureidoamidi* ***4a-o***

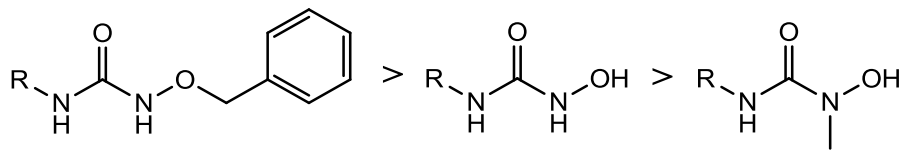
Citostatsko djelovanje ureidoamida **4a**-**o** ispitano je *in vitro* na sljedeće maligne stanične linije: karcinom dojke (MCF-7), kolorektalni adenokarcinom (SW620), karcinom debelog crijeva (HCT 116), akutna limfoblastna leukemija (MOLT-4) i karcinom pluća (H460) (Perković *et al*, 2010).

Antitumorska ispitivanja su pokazala da ureidoamidi nisu značajno inhibirali rast ispitanih stanica u najvećoj ispitanoj koncentraciji ($IC_{50} \geq 100$ μmol L^{-1}).

3.2.1.2. *N- i O-Supstituirane hidroksiuree* **6a-l**

Citostatsko djelovanje hidroksiurea **6a**-**l** ispitano je *in vitro* na sljedeće maligne stanične linije: karcinom grlića maternice (HeLa), MCF-7, karcinom gušterače (MiaPaCa-2), karcinom prostate (PC-3), SW620, karcinom stanica jetre (Hep G2), leukemija (L1210), zloćudno preobraženi T-limfociti (Molt4/C8 i CEM) i na staničnu liniju normalnih fibroblasta porijeklom iz čovjeka (WI 38) (Tablica 25) (Perković *et al*, 2008). Iz rezultata je vidljivo da su općenito bolje djelovanje na sve stanične linije pokazali *O*-benzilni derivati (**6a**,**c**,**e**,**g**,**i**,**k**) u usporedbi s derivatima sa slobodnom hidroksilnom skupinom (*N*-metil derivati **6b**,**d**,**f**,**h**,**j**,**l** i nesupstituirane hidroksiuree sintetizirane u prijašnjim istraživanjima).

Usporedbom hidroksiurea **6** i nesupstituiranih ranije sintetiziranih i ispitanih hidroksiurea (Opačić *et al*, 2005) koje su sve derivati istih aminokiselina (L-leucina, D-fenilglicina i L-fenilalanina) može se zaključiti da derivati D-fenilglicina i L-fenilalanina imaju snažnije citostatsko djelovanje od derivata L-leucina i da njihovi *N*-metilni derivati imaju najslabije djelovanje uz nekoliko iznimaka koje će biti spomenute kasnije (Slika 12).



Slika 12. Biološko djelovanje hidroksiurea s obzirom na supstituent na terminalnom dušiku.

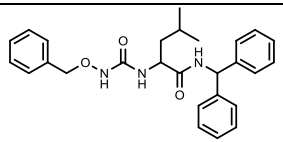
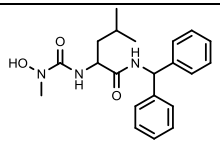
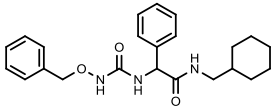
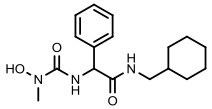
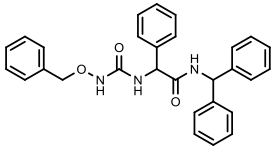
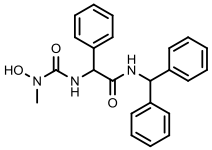
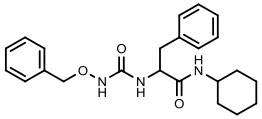
Ako se promatraju *O*-benzilni derivati **6** s benzhidrilnim amidnim supstituentom može se uočiti da aktivnost raste s obzirom na aminokiselinu prisutnu u molekuli na sljedeći način: L-leucin < D-fenilglicin < L-fenilalanin za sve stanične linije. To pravilo vrijedi i ako se promatraju *N*-metilni derivati s benzhidrilnim amidnim supstituentom, uz nekoliko već spomenutih iznimaka.

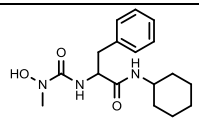
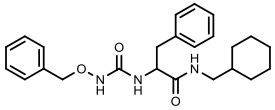
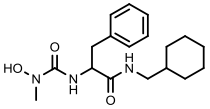
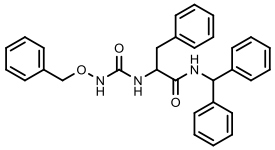
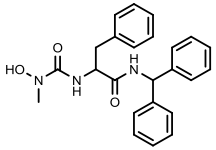
Najjače citostatsko djelovanje ($IC_{50} \leq 1$ μmol L^{-1}) na dvije stanične linije, SW620 i/ili Hep G2, pokazala su tri *N*-metilna derivata, **6d**,**f**,**l** od kojih je najbolje selektivno djelovanje pokazao derivat **6l** i to na staničnu liniju SW 620 (IC_{50} = 0,1 μmol L^{-1}). Od *O*-benzilnih derivata, najjače citostatsko djelovanje ($IC_{50} \leq 1$ μmol L^{-1}), također na stanične linije SW620 i/ili Hep G2, pokazali su spojevi **6e** i **6k**. Od navedenih spojeva derivat **6l** osim najjačeg djelovanja na staničnu liniju SW620 ima i najslabije djelovanje na staničnu liniju normalnih fibroblasta WI 38 (IC_{50} = 44 μmol L^{-1}) što ukazuje na njegovu manju toksičnost na zdrave stanice.

Tri *N*-metilna derivata (**6b**,**h**,**j**) bila su gotovo potpuno inaktivna u najvećoj ispitanoj koncentraciji (IC_{50} > 100 μmol L^{-1}) na sve testirane stanične linije. Većina *O*-benzilnih derivata (**6a**,**c**,**e**,**k**) pokazali su izraženo citostatsko djelovanje na najveći broj testiranih staničnih linija (IC_{50} = 0,8 do 48,1 μmol L^{-1}); najjače na HeLa stanice (IC_{50} = 14,4 do 28,6 μmol L^{-1}), na MCF-7 stanice (**6c**,**e**,**k**, IC_{50} = 4,9 do 9,6 μmol L^{-1}), na MIA PaCa-2 stanice (**6c**,**k**, IC_{50} = 11,8 i 22,1 μmol L^{-1}), na PC-3 stanice (**6c**,**e**,**k**,**l**, IC_{50} = 7,4 do 19,5 μmol L^{-1}) i na limfocitne i stanične linije leukemije (**6a**,**e**,**k**, IC_{50} = 11 do 28 μmol L^{-1}). Većina istaknutih derivata sadrži benzhidrilni (**6a**,**e**,**k**) osim derivata **6c** koji sadrži cikloheksanmetilni amidni supstituent.

Rezultati inhibitornog djelovanja *N*- i *O*- supstituiranih hidroksiurea dani su u Tablici 25.

Tablica 25. Inhibitorno djelovanje *N*- i *O*- supstituiranih hidroksiurea **6a-l** na rast malignih staničnih linija i normalnih fibroblasta (WI 38)

Spoj	Struktura	Stanične linije (IC_{50}[a] /μmol L^{-1})									
		HeLa	MCF-7	MIAPaCa-2	PC-3	SW620	Hep G2	L1210	Molt4/C8	CEM	WI 38
6a		28,6	29,9	48,1	42,9	31,2	33,8	23	28	28	27,9
6b		96,7	>100	>100	>100	>100	>100	>100	>100	>100	>100
6c		24,5	9,6	11,8	19,5	6,9	8,4	39	32	42	9,2
6d		>100	>100	43,8	44,6	0,7	1	>100	>100	>100	9,3
6e		25,7	5,9	38,4	15,2	0,8	4,6	12	14	28	20,1
6f		>100	63,4	>100	45,7	0,7	65,9	77	66	92	31,5
6g		49,6	40,9	78,1	52,2	53	27,1	44	45	51	47,1

6h		>100	>100	>100	>100	>100	>100	>100	>100	>100	>100
6i		54,1	77,4	59,9	>100	92,2	>100	73	98	135	68,4
6j		>100	>100	>100	>100	>100	>100	>100	>100	>100	>100
6k		14,4	4,9	22,1	7,8	1	0,9	11	12	12	8,1
6l		39,8	>100	50,9	7,4	0,1	24,8	61	57	89	44

[a] IC_{50} – koncentracija spoja kod koje je rast stanica inhibiran za 50 %.

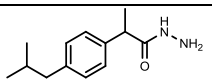
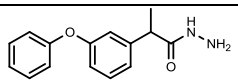
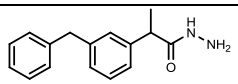
3.2.1.3. *4-Benziloksisemikarbazid (**8**) i 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazid* (***9***)

Citostatsko djelovanje spojeva **8** i **9** ispitano je *in vitro* na sljedeće maligne stanične linije: L1210, CEM, HeLa, HCT 116, MCF-7 i H460. Navedeni spojevi predstavljaju prekursore u sintezi semikarbazidnih derivata NSAID te prema očekivanjima nisu značajno inhibirali rast ispitanih stanica u najvećoj ispitanoj koncentraciji (IC_{50} > 250 μmol L^{-1} za stanične linije L1210, CEM i HeLa; IC_{50} > 100 μmol L^{-1} za stanične linije HCT 116, MCF-7 i H460).

3.2.1.4. *Hidrazidi NSAID **11a-c***

Citostatsko djelovanje spojeva **11a-c** ispitano je *in vitro* na sljedeće maligne stanične linije: L1210, CEM, HeLa, HCT 116, MCF-7 i H460. Navedeni spojevi predstavljaju prekursore u sintezi semikarbazidnih derivata NSAID te su pokazali vrlo slabo antitumorsko djelovanje. Rezultati inhibitornog djelovanja hidrazida dani su u Tablici 26.

Tablica 26. Inhibitorno djelovanje hidrazida **11a-c** na rast malignih staničnih linija.

Spoj	Struktura	Stanične linije (IC_{50}[a] /μmol L^{-1})					
		L1210	CEM	HeLa	HCT 116	MCF-7	H460
11a		170±26	147±22	129±24	>100	>100	>100
11b		212±9	154±6	120±4	>100	>100	>100
11c		212±26	150±23	142±17	>100	>100	>100

[a] IC_{50} – koncentracija spoja kod koje je rast stanica inhibiran za 50 %.

3.2.1.5. *1-Acil-4-cikloalkil, 1-acil-4-aril i 1-acil-4-hidroksi semikarbazidi NSAID **13a-y***

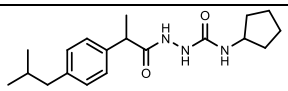
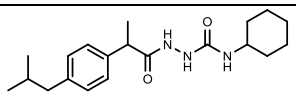
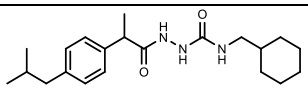
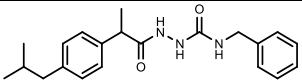
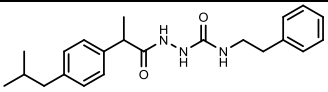
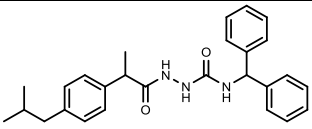
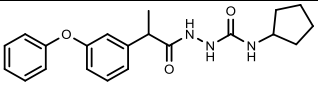
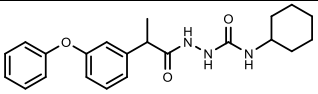
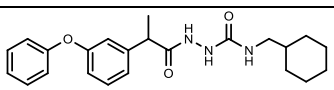
Citostatsko djelovanje spojeva **13a-y** ispitano je *in vitro* na sljedeće maligne stanične linije: L1210, CEM, HeLa, HCT 116, MCF-7 i H460. Većina ispitanih derivata pokazala je najjače djelovanje na MCF-7 stanice što se može pripisati i njihovoj jačoj osjetljivosti u odnosu na druge stanične linije.

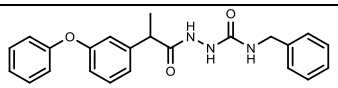
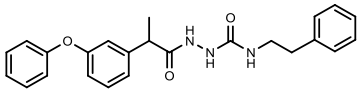
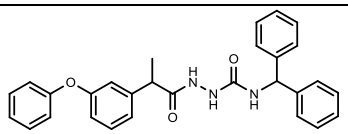
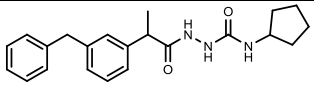
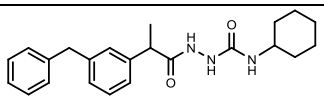
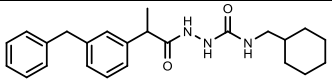
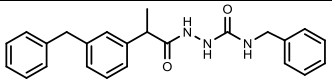
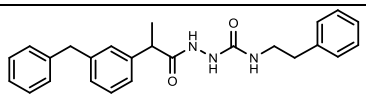
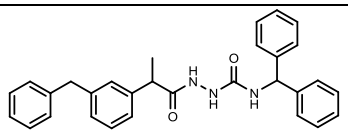
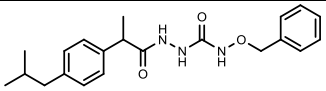
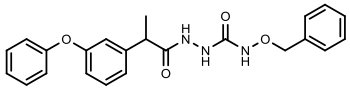
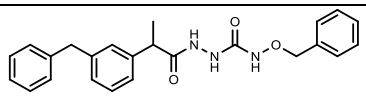
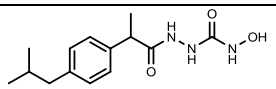
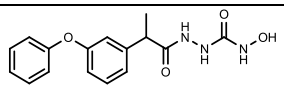
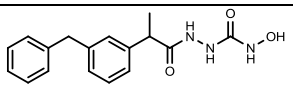
Najjače djelovanje pokazali su derivati **13f** (derivat ibuprofena), **13l** (derivat fenoprofena) i **13s** (derivat reduciranog ketoprofena) koji svi imaju benzhidrilni supstituent (IC_{50} = 3 do 23

μmol L^{-1}). Ako promatramo derivate s istim bočnim lancem (cikloalkilni ili arilni supstituent) aktivnost raste na sljedeći način: ibuprofen < reducirani ketoprofen < fenoprofen. Općenito alifatski derivati su pokazali nešto slabije djelovanje u odnosu na aromatske. Najslabije djelovanje u seriji **13** pokazali su *O*-benzilni i derivati sa slobodnom hidroksilnom skupinom (**13t-y**) te su bili ili slabo aktivni ili inaktivni u najvećoj ispitanoj koncentraciji. Upravo za te derivate se očekivalo najbolje rezultate s obzirom da sadrže hidroksamsku kiselinu u strukturi. Međutim, spojevi s lipofilnim benzidrilnim supstituentom dali su najbolje rezultate.

Rezultati inhibitornog djelovanja semikarbazida dani su u Tablici 27.

Tablica 27. Inhibitorno djelovanje semikarbazida **13a-y** na rast malignih staničnih linija

Spoj	**Struktura**	**Stanične linije (IC_{50}[a] /μmol L^{-1})**					
		L1210	**CEM**	**HeLa**	**HCT 116**	**MCF-7**	**H460**
13a		103±62	74±22	80±44	32±4	18±1	32±8
13b		30±7	53±6	69±45	28±0.2	19±3	35±0.01
13c		44±29	26±6	142±109	21±7	13±0.7	29±2
13d		110±10	74±18	173±8	34±1	29±12	59±11
13e		180±12	148±45	≥250	26±2	23±5	51±14
13f		20±0	23±4	22±12	13±0.06	6±1	16±1
13g		55±16	73±27	39±20	36±20	20±5	33±17
13h		35±7	53±8	48±5	22±7	17±2	24±2
13i		22±1	24±1	24±0	15±0.1	13±0.5	19±1

13j		63±1	72±14	70±14	27±0.2	17±1	19±13
13k		53±1	71±9	64±8	18±1	19±4	22±1
13l		15±1	11±7	5.7±0.2	5±0.7	7±3	3±0.4
13m		69±12	66±4	43±12	33±8	19±2	38±2
13n		25±1	25±5	45±12	22±1	16±3	23±0.4
13o		22±1	20±1	20±0	16±3	18±10	18±0.5
13p		31±3	49±6	94±42	24±0.4	12±8	28±2
13r		31±3	29±3	58±7	20±5	15±0.8	22±2
13s		19±0	15±0	5.0±0.5	8±2	5±1	4±0.6
13t		192±81	≥250	178±3	76±10	50±11	>100
13u		84±10	91±9	76±4	76±21	81±7	>100
13v		79±4	101±11	78±2	56±10	41±18	82±19
13w		108±11	205±63	160±13	>100	>100	>100
13x		125±11	183±95	≥250	>100	>100	>100
13y		113±11	155±71	176±26	>100	>100	>100

[a] IC_{50} – koncentracija spoja kod koje je rast stanica inhibiran za 50 %.

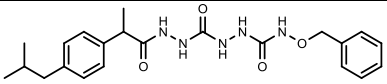
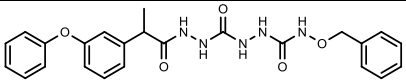
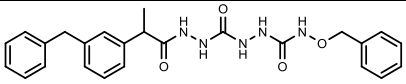
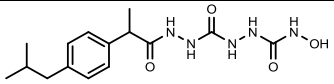
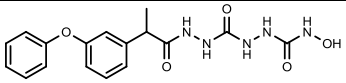
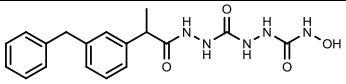
3.2.1.6. *1-Acil-5-benziloksi i 1-acil-5-hidroksikarbamoil karbazidi NSAID* ***14a-f***

Citostatsko djelovanje spojeva **14a-f** ispitano je *in vitro* na sljedeće maligne stanične linije: L1210, CEM, HeLa, HCT 116, MCF-7 i H460.

Derivati **14** nisu pokazali značajnu aktivnost na ispitane stanične linije osim *O*-benzilnih (**14a-c**) na staničnu liniju MCF-7 (IC_{50} = 19 do 26 μmol L^{-1}). Derivati sa slobodnom hidroksilnom skupinom (**14d-f**) bili su inaktivni u najvećoj ispitanoj koncentraciji (IC_{50} > 250 μmol L^{-1} za stanične linije L1210, CEM i HeLa; IC_{50} > 100 μmol L^{-1} za stanične linije HCT 116, MCF-7 i H460).

Rezultati inhibitornog djelovanja derivata **14a-f** dani su u Tablici 28.

Tablica 28. Inhibitorno djelovanje 1-acil karbazida **14a-f** na rast malignih staničnih linija

Spoj	Struktura	Stanične linije (IC_{50}[a] /μmol L^{-1})					
		L1210	CEM	HeLa	HCT 116	MCF-7	H460
14a		105±10	106±34	98±55	56±31	19±8	71±30
14b		103±5	90±10	88±4	84±16	26±10	>100
14c		102±1	96±3	76±20	64±21	19±10	>100
14d		>250	>250	>250	>100	>100	>100
14e		>250	>250	≥250	>100	>100	>100
14f		>250	>250	≥250	>100	>100	>100

[a] IC_{50} – koncentracija spoja kod koje je rast stanica inhibiran za 50 %.

3.2.2. ANTIVIRUSNA ISPITIVANJA

Antivirusna ispitivanja provedena su na Rega Institute for Medical Research, Katholieke Universiteit Leuven, Belgija, prema ranije opisanoj metodi (Perković *et al*, 2008).

3.2.2.1. *N- i O-Supstituirane hidroksiuree* ***6a-l***

Antivirusno djelovanje *N*- i *O*- supstituiranih hidroksiurea ispitano je na sljedeće viruse: virus humane imunodeficijencije tip 1 i 2 (HIV-1 i 2) u kulturi CEM stanica, herpes simpleks virus tip 1 (KOS) i tip 2 (G), vaccinia virus i vesicular stomatitis virus u kulturi HEL stanica, Coxsackie virus B4 i respiracijski sincicijski virus (RSV) u kulturi HeLa stanica, parainfluenza-3 virus, reovirus-1, Sindbis virus i Punta Toro virus u kulturi Vero stanica, Feline corona virus u kulturi CRFK stanica i virus influence tip A (H1N1 i H3N2 subtipovi) i tip B u kulturi MDCK stanica.

Hidroksiuree **6** nisu pokazale specifično antivirusno djelovanje, tj. minimalna efektivna koncentracija (EC_{50}, efektivna koncentracija kod koje je stvaranje plakova virusa smanjeno za 50 %) bila je veća od 1/5 minimalne citotoksične koncentracije (*MCC*, minimalna citotoksična koncentracija kod koje je mikroskopski vidljiva promjena morfologije normalne stanice, subtoksična koncentracija) (rezultati nisu pokazani).

3.2.2.2. *Derivati* **8**, **9**, **11**, **13**, **14**

Antivirusno djelovanje derivata **8**, **9**, **11**, **13** i **14** ispitano je na sljedeće viruse: HIV-1 i 2, KOS i G, vaccinia virus i vesicular stomatitis, parainfluenza-3, reovirus-1, Sindbis, Coxsackie B4, Punta Toro virus, RSV, Feline herpes i corona virus, virus influence tip A (H1N1 i H3N2 subtipovi) i tip B, varicella zoster i citomegalovirus u HEL stanicama.

Ispitani derivati nisu pokazali specifično antivirusno djelovanje tj. inhibiciju u subtoksičnim koncentracijama na sve ispitane viruse u kulturama stanica osim derivata **13a**, **g** i **m** na citomegalovirus u HEL stanicama (sojevi AD-169 i Davis). Spoj **13a** (derivat ibuprofena) imao je EC_{50} (efektivna koncentracija kod koje je stvaranje plakova virusa smanjeno za 50 %) 11,19 (AD-169) i 7,16 μmol L^{-1} (Davis); **13g** (derivat fenoprofena) 14,47 (AD-169) i 7,49 μmol L^{-1} (Davis); **13m** (derivat ketoprofena) 14,89 (AD-169) i 11,24 μmol L^{-1} (Davis). Minimalna citotoksična koncentracija (*MCC*) je za navedene derivate bila 100 μmol L^{-1} (indeks selektivnosti MCC/EC_{50} = 7–14) (rezultati nisu pokazani). Sva tri navedena derivata imaju ciklopentilni supstituent pa se može zaključit da je on bitan za antivirusno djelovanje.

3.2.3. Antimikrobna ispitivanja derivata aminokiselina 4 i 6

Antimikrobno ispitivanje provedeno je na Zavodu za mikrobiologiju Farmaceutsko-biokemijskog fakulteta u Zagrebu.

Antimikrobno i antifungalno djelovanje spojeva **4** i **6** ispitano je korištenjem difuzijske metode (bušenjem cilindrima: visine 10, vanjskog promjera 8, a unutrašnjeg 6 mm) (European Pharmacopoeia, 2006). Korištene su sljedeće vrste: *B. subtilis* (ATCC 6633), *S. aureus* (ATCC 25923), *E. faecalis* (ATCC 29212), *K. rhizophila* (ATCC 9341), *E. coli* (ATCC 10536), *S. enterica* subsp. *enterica* (ATCC 13076), *P. aeruginosa* (ATCC 27853), *C. albicans* (ATCC 10231). Niti jedan od ispitanih spojeva nije pokazao zone inhibicije rasta te su smatrani antimikrobno inaktivnima pri ispitivanoj koncentraciji (serija **4**: 20 mg mL^{-1}, serija **6**: 31,75 mg mL^{-1}) i provedenoj metodi, djelomično i zbog njihove slabe topljivosti (serija **6**) u Müller-Hinton agaru (prilikom izvođenja pokusa supstancije su se taložile, rezultati nisu prikazani). Niti jedan od ispitanih spojeva nije pokazao antifungalno djelovanje.

Minimalna inhibitorna (*MIC*) i minimalna mikrobicidna koncentracija (*MMcC*) određene su metodom serijalnog mikrorazrijeđenja (CLSI) (NCCLS, 1997).

Oksitetraciklin, norfloksacin i nistatin su korišteni kao kontrola osjetljivosti mikrobnih kultura u metodi difuzije.

3.2.4. Antioksidativno djelovanje, inhibicija lipidne peroksidacije linolne kiseline i inhibicija lipooksigenaze

Ispitivanje antioksidativnog djelovanja, inhibicije lipidne peroksidacije linolne kiseline i inhibicija lipooksigenaze provedena su na Department of Pharmaceutical Chemistry, School of Pharmacy, Aristotles University of Thessaloniki, Solun, Grčka prema ranije opisanoj metodi (Pontiki *et al*, 2007; Re *et al*, 1999; Taraborewala *et al*, 1990).

3.2.4.1. *Ureidoamidi **4a-o***

Ispitivanje antioksidativnog djelovanja temelji se na mjerenju stupnja interakcije supstancija sa stabilnim slobodnim radikalom 1,1-difenil-2-pikrilhidrazil radikalom (DPPH) i

opisuje njihovu sposobnost hvatanja slobodnih radikala u sustavu bez željeza što pokazuje kolika je njihova reducirajuća aktivnost.

Na temelju te činjenice ispitana je interakcija ureidoamida **4** s DPPH, prema ranije opisanoj metodi (Pontiki *et al*, 2007) mjerenjem apsorbancije ispitivanih otopina nakon 20 i 60 min kod dvije koncentracije ispitivanih spojeva (0,05 i 0,1 mmol L^{-1}), a rezultati su prikazani u Tablici 29. Rezultati pokazuju da je interakcija s DPPH bila vrlo slaba (ispod 10 % za sve ispitane spojeve) te da nije došlo do promjene u jačini interakcije ovisno o koncentraciji ispitivanih derivata i vremenu.

Princip ispitivanja stupnja inhibicije lipidne peroksidacije linolne kiseline koristi sposobnost azo spojeva da generiraju slobodne radikale spontanom termičkom razgradnjom (koriste se za *in vitro* studije nastajanja slobodnih radikala). U ovim pokusima korišten je vodotopljivi 2,2'-azobis(2-amidinopropan) dihidroklorid (AAPH) kao inicijator slobodnih radikala, koji oksidiraju linolnu kiselinu do konjugiranog peroksidnog diena (Re *et al*, 1999).

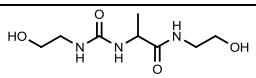
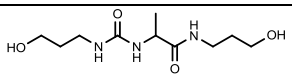
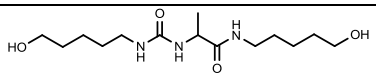
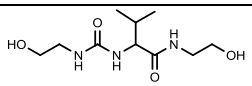
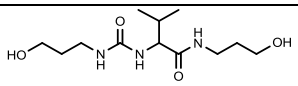
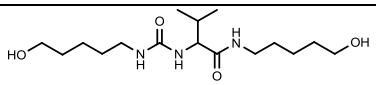
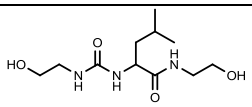
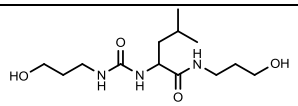
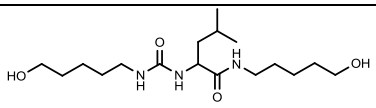
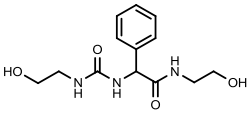
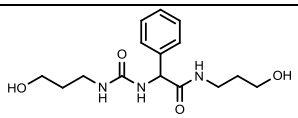
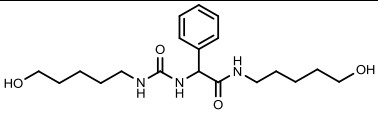
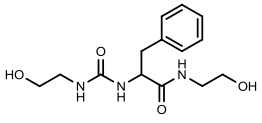
Rezultati ispitivanja sposobnosti inhibicije lipidne peroksidacije linolne kiseline pokazuju slabu do umjerenu inhibiciju lipidne peroksidacije linolne kiseline za većinu ispitanih derivata. Iznimka su derivati **4c**,**m**,**g**,**j** (vrijednosti od 59,6–94,6 %, Tablica 29).

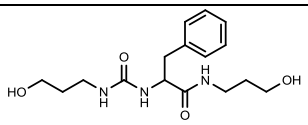
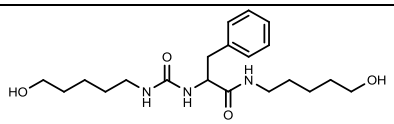
Lipooksigenaze (LO) oksidiraju masne kiseline koje u svojoj strukturi sadrže konjugiranu triensku strukturu na specifičnim položajima, do hidroperoksida, prekursora leukotriena (medijatori upale). Većina inhibitora lipooksigenaze su antioksidansi ili hvatači slobodnih radikala, zbog toga što se lipooksigenacija događa preko ugljikovih radikala.

Sposobnost inhibicije LO ispitana je enzimskim testom temeljenim na mjerenju UV apsorpcije (Tablica 29) (Pontiki *et al*, 2007; Taraborewala *et al*, 1990). Ureidoamidi su umjereno inhibirali LO što pokazuju vrijednosti IC_{50} izmjerene za derivate **4b**, **4d**, **4g**, **4j**, **4k**, **4m** (88–100 μmol L^{-1}).

Kao poredbene tvari korišteni su antioksidansi kavena kiselina, nordihidrogvajaretična kiselina (NDGA) i troloks.

Tablica 29. Interakcija s DPPH, inhibicija lipidne peroksidacije linolne kiseline i *in vitro* inhibicija LO ureidoamida **4**

Spoj	Struktura	DPPH 20 min[a] (%)	DPPH 60 min[a] (%)	DPPH 20 min[b] (%)	DPPH 60 min[b] (%)	Inhibicija LP[b] (%)	Inhibicija LO[b]
4a		1,7	0,5	n.a.[c]	n.a.	n.a.	28,1
4b		n.a.	0,9	0,3	1	32,9	49,5[d]
4c		3,2	3,9	1,4	4,8	94,6	22,6
4d		3,4	6,3	2,1	3,5	24,2	55,7[d]
4e		2,7	6,3	5,0	4,7	15,0	37,8
4f		n.a.	1,2	2,3	5,2	43,7	36,1
4g		1,2	0,9	n.a.	1,5	59,6	52,8[d]
4h		n.a.	3,2	0,9	3,6	16,1	42,6
4i		1,2	1,1	2,7	2,8	11,1	42,5
4j		2,3	3,8	5,7	7,5	63,1	50,1[d]
4k		n.a.	2,2	4,9	1,3	47,9	55,6[d]
4l		1,6	2,4	0,9	3,7	3,9	36,9
4m		0,5	3,0	n.a.	1,1	87,4	49,2[d]

4n		0,9	1,8	n.a.	0,9	19,9	39,7
4o		1,8	3,3	2,5	3,7	16,6	27,6
Kavena kiselina		n.o.[e]	n.o.	n.o.	n.o.	n.o.	600
NDGA		81	83	93	97	n.o.	n.o.
Troloks		n.o.	n.o.	n.o.	n.o.	63	n.o.

Koncentracije: [a] 5×10^{-5} mol L^{-1}; [b] 1×10^{-4} mol L^{-1}; [c] n.a. – nema aktivnosti; [d] IC_{50} (μmol L^{-1}) su određene: 100 (**4b**), 92,5 (**4d**), 98 (**4g**), 100 (**4j**), 88 (**4k**), 100 (**4m**); [e] n.o. – nije određeno.

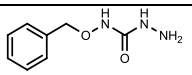
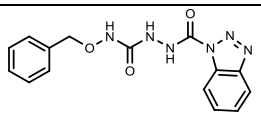
3.2.4.2. *Derivati **8** i **9***

Interakcija derivata **8** i **9** s DPPH promatrana je kod jedne koncentracije spojeva (0,1 mmol L^{-1}) u ovisnosti o vremenu (nakon 20 i 60 min). Rezultati su pokazali da spoj **9** stupa u snažnu interakciju s DPPH (85 %) za razliku od spoja **8** (14 %). Nije bilo značajnih promjena u interakciji spojeva s DPPH u ovisnosti o vremenu.

Rezultati ispitivanja inhibicije lipidne peroksidacije linolne kiseline pokazali su slabu aktivnost za oba derivata (36 i 31 %) (Tablica 30).

Sposobnost inhibicije LO derivatima **8** i **9** ispitana enzimskim testom temeljenim na mjerenju UV apsorpcije polazala je inaktivnost oba derivata za danu koncentraciju.

Tablica 30. Teorijski izračunate clog *P* vrijednosti, interakcija s DPPH, inhibicija lipidne peroksidacije linolne kiseline i *in vitro* inhibicija lipooksigenaze derivata **8** i **9**

Spoj	**Struktura**	**clog *P*[a]**	**DPPH 20 min[b] (%)**	**DPPH 60 min[b] (%)**	**Inhibicija LP (%)**	**Inhibicija LO[c]**
8		0,27	6	14	36	n.a.[d]
9		0,27	6	14	36	n.a.
NDGA		n.o.[e]	80	91	n.o.	84[f]
Troloks		n.o.	n.o.	n.o.	63	n.o.

[a] Lipofilnost molekule teorijski je izračunata kao clog *P* u smjesi *n*-oktanola i pufera CLOGP programom (Biobyte Corp). Koncentracije: [b] 5×10^{-5} mol L^{-1}; [c]IC_{50} (μmol L^{-1}); [d]n.a. – nema aktivnosti; [e] n.o. – nije određeno; [f] IC_{50} = 28 μM

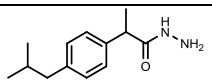
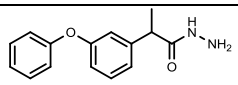
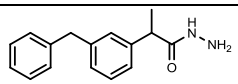
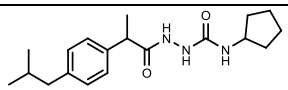
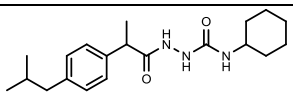
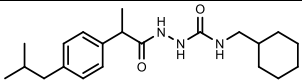
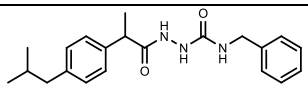
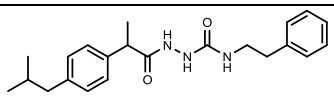
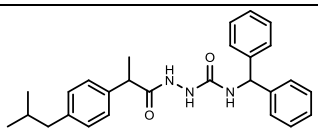
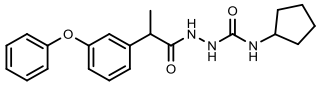
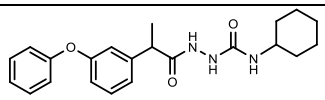
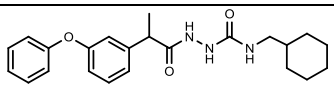
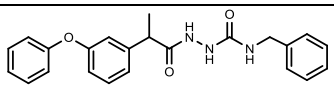
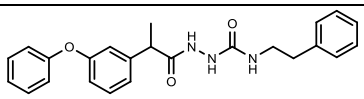
3.2.4.3. *NSAID derivati* **11**, **13** *i* **14**

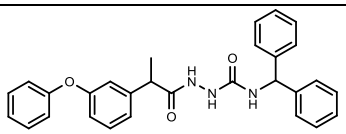
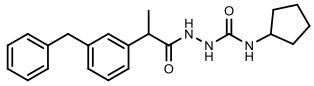
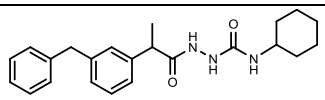
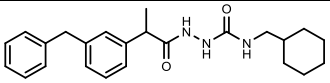
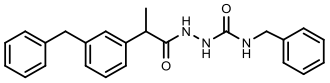
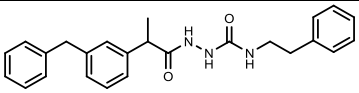
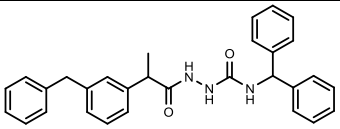
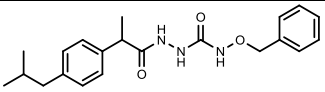
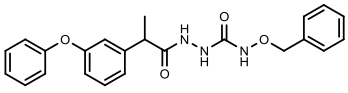
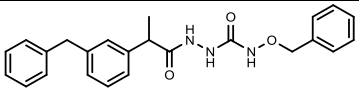
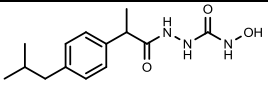
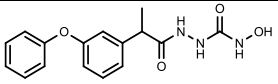
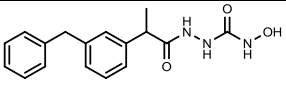
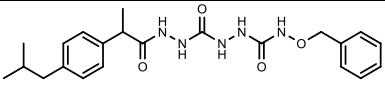
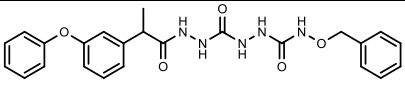
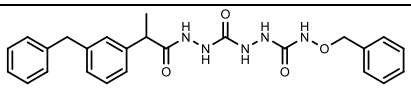
Interakcija NSAID derivata **11**, **13** i **14** s DPPH promatrana je kod koncentracije spojeva 0,1 mmol L^{-1} u ovisnosti o vremenu (nakon 20 i 60 min). Rezultati su pokazali da hidrazidi NSAID **11** ne stupaju u interakciju s DPPH, semkarbazidi **13a-s** stupaju u vrlo slabu interakciju s DPPH (najbolja interakcija uočena je za fenoprofenski derivat **5j**, 24 %) i aktivnost pada s obzirom na NSAID u molekuli (fenoprofen > reducirani ketoprofen > ibuprofen). *O*-Benzilni derivati **13t-v** također ne stupaju u interakciju s DPPH za razliku od derivata sa slobodnom hidroksilnom skupinom **13w-y** koji su pokazali nabolju interakciju koja je rasla u ovisnosti o vremenu (54–66 %). Lipofilnost spojeva ne utječe na njihovu interakciju s DPPH. Interakcija svih ispitanih spojeva s DPPH slabija je od interakcije NDGA s DPPH.

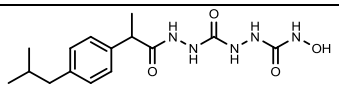
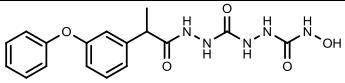
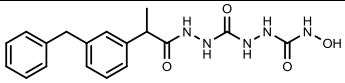
Nekoliko derivata pokazalo je vrlo snažnu inhibiciju lipidne peroksidacije linolne kiseline (99–100 %) i to fenoprofenski derivat **13l**, ibuprofenski derivat **13t** i ketoprofenski derivat **14c**. Osim njih još su neki derivati pokazali jaku inhibiciju, **13f**, **13r** i **13s** (71–88 %). Općenito, bolje djelovanje su pokazali derivati s aromatskim supstituentom od onih s cikloalkilnim. Derivati s benzhidrilnim supstituentom pokazali su jako dobro djelovanje koje se može korelirati s njihovim citostatskim djelovanjem. Za razliku od toga, derivati koji su najbolje inhibirali lipidnu peroksidaciju linolne kiseline vrlo su slabo stupali u interakciju s DPPH.

Sposobnost inhibicije LO derivatima **11**, **13** i **14** ispitana je enzimskim testom temeljenim na mjerenju UV apsorpcije (Tablica 31) (Pontiki *et al*, 2007; Taraborewala *et al*, 1990). Najsnažniji inhibitori bili su **13s**, **13l** i **13j**, (68–96 %), ponovo spojevi s benzhidrilnim supstituentom (**13s**,**l**) te s benzilnim supstituentom (**13j**). Za njih su izmjerene i pripadajuće inhibitorne koncentracije (IC_{50}).

Tablica 31. Teorijski izračunate clog *P* vrijednosti, interakcija s DPPH, inhibicija lipidne peroksidacije linolne kiseline i *in vitro* inhibicija lipooksigenaze derivata **11**, **13** i **14**

Spoj	Struktura	clog *P*[a]	DPPH 20 min[b] (%)	DPPH 60 min[b] (%)	Inhibicija LP[c] (%)	Inhibicija LO[b]
11a		2,41	n.a.[g]	1	6	8
11b		2,55	n.a.	1	10	3
11c		2,52	n.a.	n.a.	6	10
13a		4,04	4	7	26	n.a.
13b		4,60	n.a.	n.a.	33	n.a.
13c		5.22	3	6	16	25
13d		4.34	n.a.	n.a.	12	n.a.
13e		4,67	n.a.	n.a.	33	n.a.
13f		6,11	n.a.	n.a.	74	46
13g		4,19	16	19	21	n.a.
13h		4,74	19	22	63	13
13i		5,36	18	21	64	12
13j		4,48	22	24	11	68[c]
13k		4,81	4	7	51	20

13l		5,83	2	4	99	81[d]
13m		4,15	4	12	n.a.	n.a.
13n		4,71	3	10	31	10
13o		5,33	5	12	61	9
13p		4,45	3	9	48	n.a.
13r		4,78	8	15	71	17
13s		5,80	4	9	88	96[e]
13t		4,09	n.a.	n.a.	100	n.a.
13u		4,24	n.a.	n.a.	8	6
13v		4,20	3	6	n.a.	n.a.
13w		1,57	29	54	20	7
13x		1.71	39	66	22	3
13y		1,68	36	59	8	8
14a		2,93	11	18	13	n.a.
14b		3,07	8	15	26	n.a.
14c		3,04	10	19	100	n.a.

14d		0,41	24	46	15	n.a.
14e		0,55	23	48	13	n.a.
14f		0,52	28	53	28	1
NDGA		n.o.[h]	80	91	n.o.	84[e]
Troloks		n.o.	n.o.	n.o.	63	n.o.

[a] Lipofilnost molekule teorijski je izračunata kao clog *P* u smjesi *n*-oktanola i pufera CLOGP programom (Biobyte Corp). Koncentracije: [b] 1×10^{-4} mol L^{-1}; [c] IC_{50} = 75 μmol L^{-1}; [d] IC_{50} = 60 μmol L^{-1}; [e] IC_{50} = 51,5 μmol L^{-1}; [f] IC_{50} = 28 μmol L^{-1}; [g] n.a. – nema aktivnosti; [h] n.o. – nije određeno.

4. ZAKLJUČCI

1. U ovom doktorskom radu ukupno je sintetizirano 94 spoja, od čega je 68 novih, do sada nepoznatih spojeva. Strukture novih spojeva karakterizirane su uobičajenim analitičkim i spektroskopskim metodama (talište, elementarna analiza, IR, ^{1}H i ^{13}C NMR).

2. Sintetizirani su derivati aminokiselina: *N*-Btc-aminokiseline **2**, njihovi kloridi **3**, ureidoamidi **4**, amidi **5** i *N*- i *O*- supstituirane hidroksiuree **6**. Novi spojevi su svi ureidoamidi **4** koji su pripravljeni aminolizom iz odgovarajućih klorida **3** i aminoalkohola i sve *N*- i *O*- supstituirane hidroksiuree **6** koje su pripravljene aminolizom iz odgovarajućih amida **5** i hidroksilamina.

3. Sintetizirani su prekursori u sintezi semikarbazidnih derivata NSAID:
    - 1-Karbamoilbenzotriazoli (amidi 1-benzotriazolkarboksilne kiseline, aktivne uree) **7** sintetizirani su u reakcijama BtcCl-a (**1**) i amina. Korišteni su u sintezi semikarbazidnih derivata NSAID **13**. Novi spojevi su **7a**,**c**,**d** i **f**.
    - Iz 4-benziloksisemikarbazida (**8**) dobiven je 1-(1-benzotriazolkarbonil)-4-benzil-oksisemikarbazid (**9**), prekursor za sintezu 1-acil-5-benziloksi/hidroksikarbamoil-karbazida NSAID **14**. Spoj **9** se pokazao kao pogodan reagens za uvođenje acil semikarbazidne skupine u molekulu. U reakcijama sa slobodnom amino skupinom NSAID hidrazida reagirao je u dobrim iskorištenjima.

4. Sintetizirani su derivati NSAID:
    - NSAID benzotriazolidi **10** bili su prekursori u sintezi hidrazida **11** i estera **12**. Hidrazidi **11** korišteni su u sintezi semikarbazidnih derivata NSAID **13** i **14**. Dobiveni su reakcijom nukleofilne supstitucije u kojoj je benzotriazol u NSAID benzotriazolidima **10a-c** zamijenjen hidrazinom. Novi spojevi su **11b** i **c**.
    - Sintetizirani su novi esterski derivati ketoprofena **12**, koji su ujedno i aminokiselinski derivati (u reakciji nukleofilne supstitucije benzotriazol u benzotriazolidu ketoprofena **10d** zamijenjen je alkoholnom skupinom iz ureidoamida **4b**,**i**,**j**,**m**). Dobiveni spojevi su dvojni lijekovi jer sadrže dvije molekule lijeka (ketoprofena).
    - 1-Acilsemikarbazidi NSAID **13a-v** sintetizirani su iz 1-karbamoilbenzotriazola **7** i hidrazida NSAID **11** reakcijom u talini. Spojevi **13w-y** sintetizirani su katalitičkim hidrogeniranjem *O*-benzilnih derivata **13t-v**, uz katalizator Pd/C. Svi spojevi su novi.

- 1-Acil-5-benziloksikarbamoilkarbazidi NSAID **14a-c** sintetizirani su iz 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazida (**9**) i hidrazida NSAID **11** reakcijama u talini. Spojevi **14d-f** pripravljeni su katalitičkim hidrogeniranjem *O*-benzilnih derivata **14a-c** uz katalizator Pd/C. Svi spojevi su novi.

5. Provedena su sljedeća biološka ispitivanja: antitumorsko, antimikrobno, antivirusno i antioksidativno djelovanje, inhibicija lipooksigenaze i lipidne peroksidacije.
    - Antitumorsko djelovanje ispitano je za derivate **4**, **6**, **8**, **9**, **11**, **13** i **14**. Hidroksiuree **6** su pokazale značajno bolje djelovanje od ureidoamida **4** i derivata **8** i **9** koji su bili inaktivni u ispitanim koncentracijama i derivata NSAID **11**, **13** i 1**4** koji su bili slabo aktivni (najbolje djelovanje imali su benzhidrilni derivati **13f**,**l**,**s** za staničnu liniju MCF-7 (IC_{50} < 10 μmol L^{-1}). Za staničnu liniju SW620 sljedeći derivati imali su najsnažnije djelovanje: IC_{50} (**6l**) = 0,1 μmol L^{-1}; IC_{50} (**6f** i **d**) = 0,7 μmol L^{-1}; IC_{50} (**6e**) = 0,8 μmol L^{-1}. Spoj **6k** pokazao je najjači učinak na rast stanične linije Hep G2 (IC_{50} = 0,9 μmol L^{-1}).
    - Antimikrobno djelovanje ispitano je za derivate **4** i **6**. Svi ispitani spojevi bili su ili inaktivni ili vrlo slabog djelovanja.
    - Antivirusno djelovanje ispitano je za derivate **6**, **8**, **9**, **11**, **13** i **14**. Djelovanje svih ispitanih spojeva bilo je slabo. Iznimka su ciklopentilni derivati NSAID **13a**,**g**,**m** koji su imali relativno dobar indeks selektivnosti (MCC/EC_{50} = 7-14) za citomegalovirus u kulturi HEL stanica.
    - Antioksidativno djelovanje ispitano je za derivate **4**, **8**, **9**, **11**, **13** i **14**. Najbolje antioksidativno djelovanje pokazao je benzotriazolid **9** (interakcija s DPPH 85 %). Svi ostali derivati bili su slabo aktivni ili inaktivni.
    - Inhibicija lipooksigenaze i lipidne peroksidacije linolne kiseline ispitana je za derivate **4**, **8**, **9**, **11**, **13** i **14**, koji su pokazali umjerenu inhibiciju LO i LP. Najsnažniji inhibitor LO bio je benzhidrilni derivat reduciranog ketoprofena **13s** (IC_{50} = 51,5 μmol L^{-1}). Najsnažniji inhibitor lipidne peroksidacije bili su ibuprofenski derivati **13t** (*O*-benzilhidroksisemikarbazid) i **14c** (1-acil-5-hidroksikarba-moilkarbazid) (100 % inhibicije) i fenoprofenski derivat **13l** (benzhidrilni supstituent) (99 % inhibicije).

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6. ŽIVOTOPIS

Ivana Perković rođena je u Zagrebu, 03. kolovoza 1982. godine. U Zagrebu je završila osnovnu školu i V. gimnaziju. Od 2000. do 2005. godine. studirala je na Farmaceutsko-biokemijskom fakultetu u Zagrebu, studij Farmacije. Tijekom studija primala je stipendiju Ministarstva znanosti i tehnologije RH dodjeljenu za prvo mjesto na klasifikacijskom ispitu. Diplomski rad pod nazivom *Priprava polimer-lijek konjugata diklofenaka i fenoprofena* izradila je na Zavodu za farmaceutsku kemiju Farmaceutsko-biokemijskog fakulteta pod vodstvom prof. dr. sc. Branke Zorc te je diplomirala 8. rujna 2005.

Od 1.12.2005. zaposlena je na Farmaceutsko-biokemijskom fakultetu prvo kao asistent na zamjeni, a od 1.6.2009. kao asistent-znanstveni novak. Uz znanstveni rad sudjeluje u izvođenju nastave na kolegijima Zavoda za farmaceutsku kemiju. Godine 2007. upisala je poslijediplomski doktorski studij Farmaceutsko-biokemijske znanosti na Farmaceutsko-biokemijskom fakultetu. Stručni ispit propisan za magistre farmacije-pripravnike pred komisijom Ministarstva zdravstva i socijalne skrbi Republike Hrvatske položila je u listopadu 2008. nakon odrađenog propisanog pripravničkog staža i dobila licencu za samostalan rad. Član je Hrvatske ljekarničke komore.

Do sada je objavila sedam znanstvenih i sedam stručnih radova, sudjelovala s priopćenjima na više domaćih i inozemnih kongresa te održala jedno predavanje.
Udana je i majka jedne djevojčice.

Znanstveni radovi (CC):

Barbarić M, Kralj M, Marjanović M, **Husnjak (Perković) I**, Pavelić K, Filipović-Grčić J, Zorc D, Zorc B. Synthesis and in vitro antitumor effect of diclofenac and fenoprofen thiolated and nonthiolated polyaspartamide-drug conjugates. *Eur J Med Chem* **42** (2007) 20–29.

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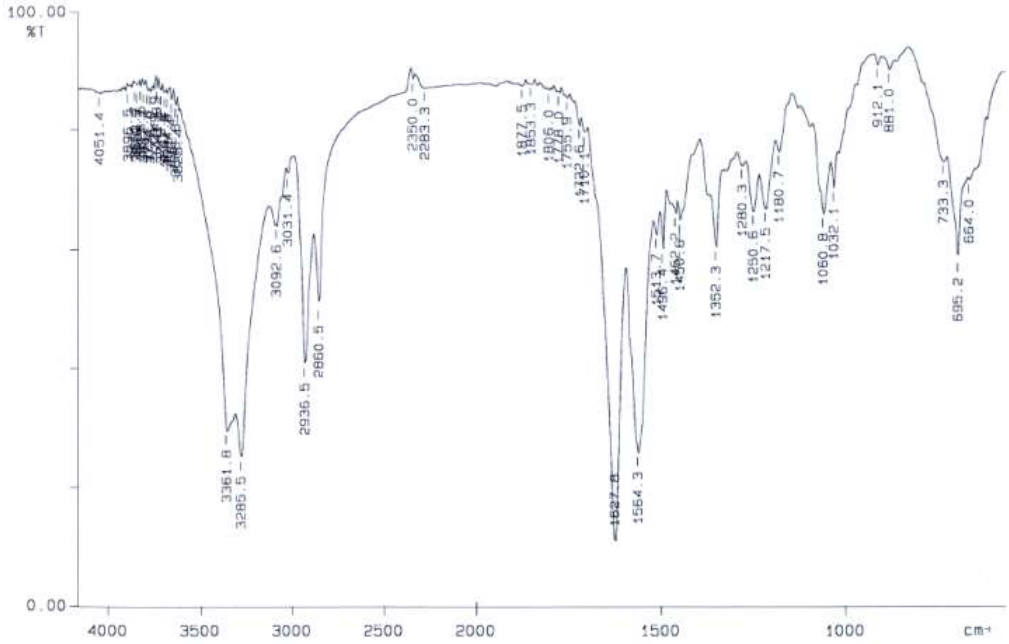
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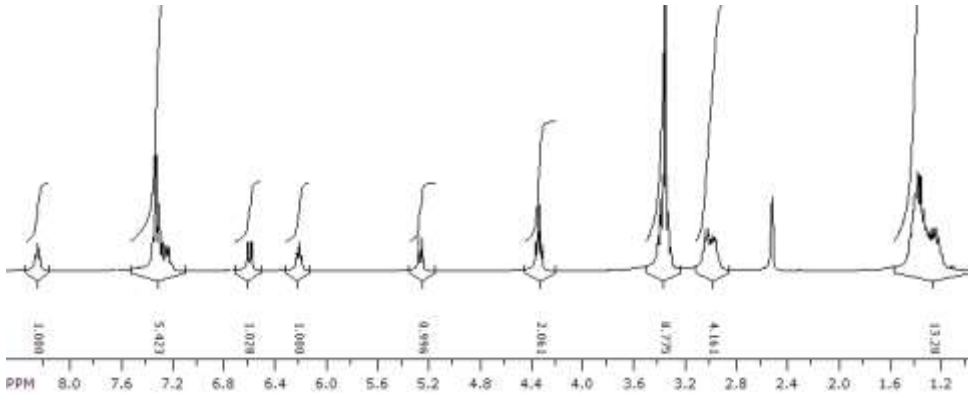
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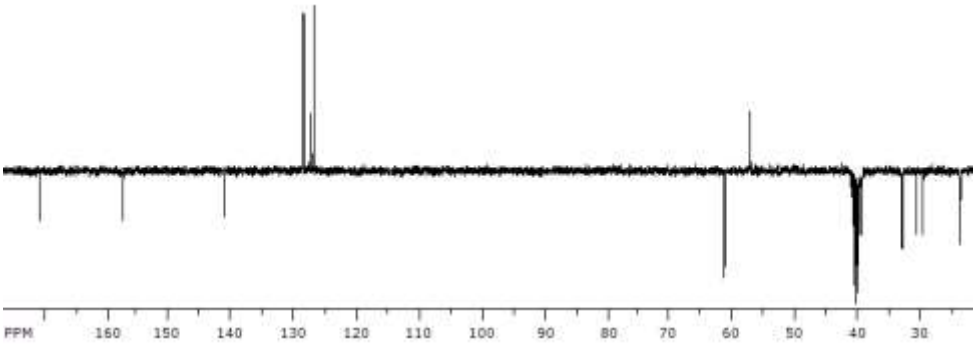
Rajić Z, Zovko Končić M, Miloloža K, **Perković I**, Butula I, Bucar F, Zorc B. Primaquine-NSAID twin drugs: Synthesis, radical scavenging, antioxidant and Fe^{2+} chelating activity. *Acta Pharm* **60** (2010) 325–337.

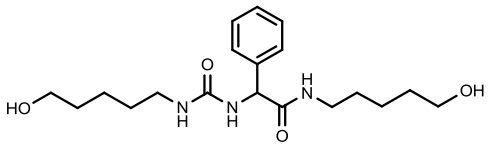
7. PRILOG A

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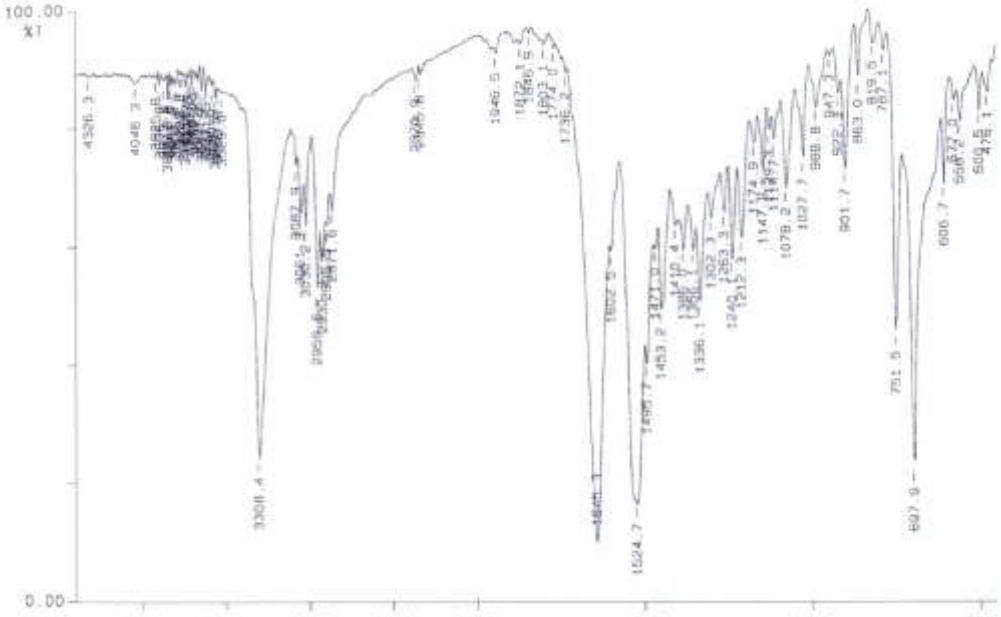

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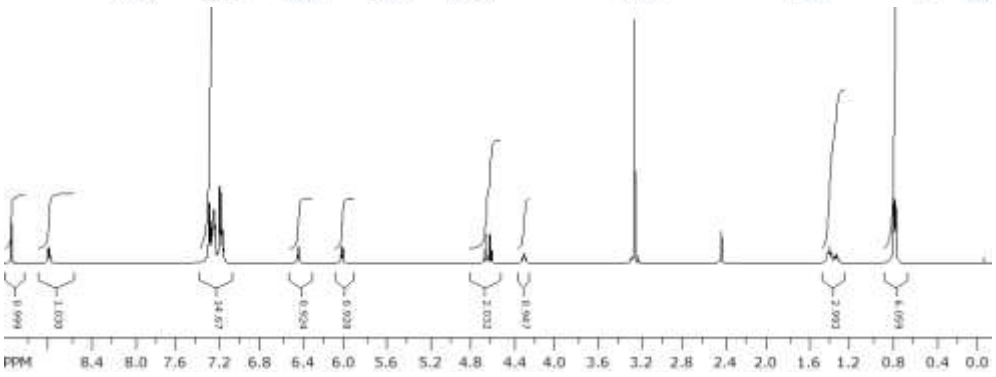

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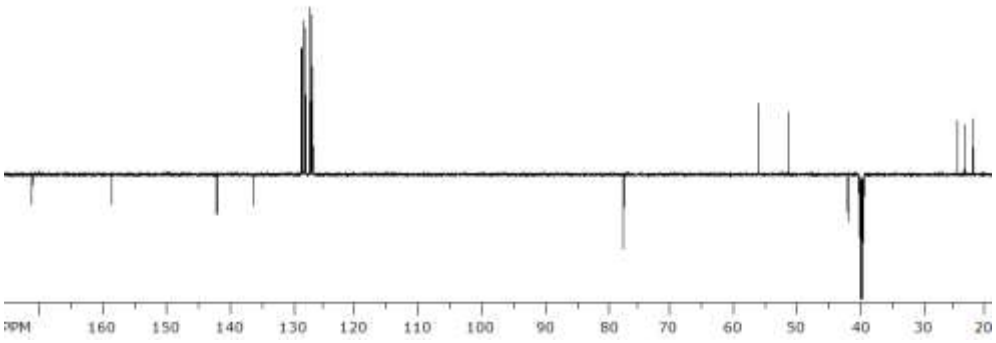

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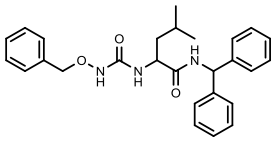

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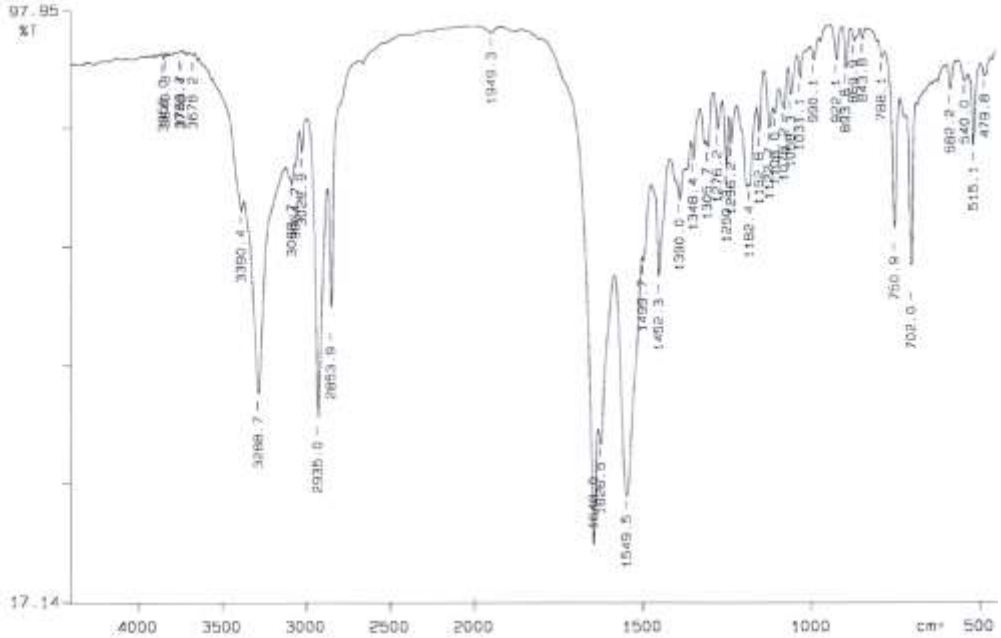

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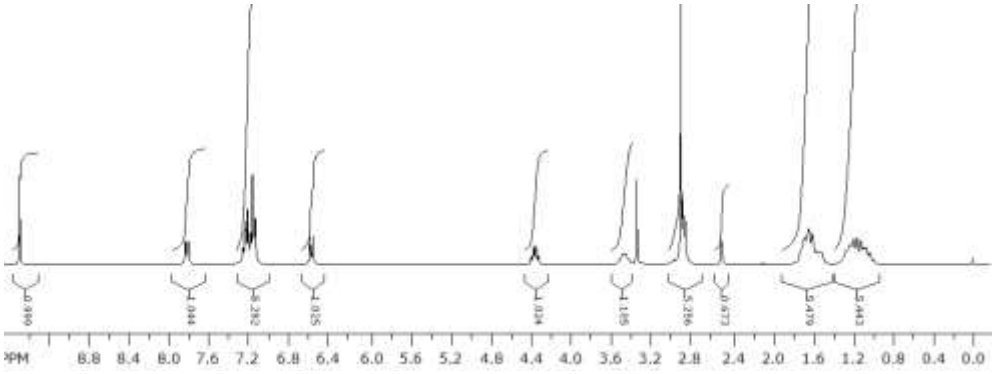

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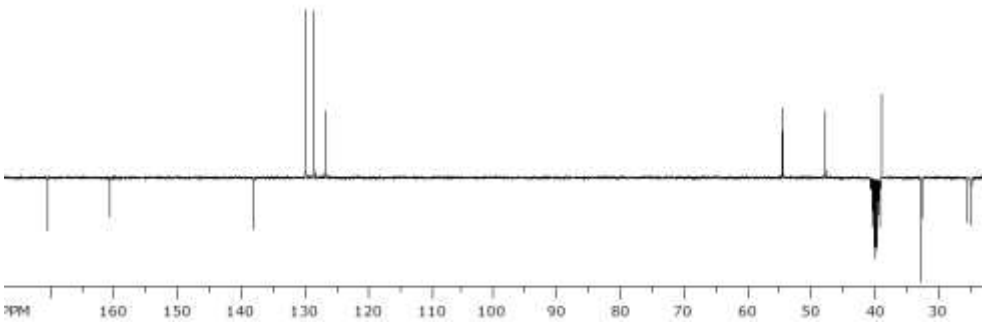

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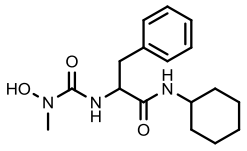


6a

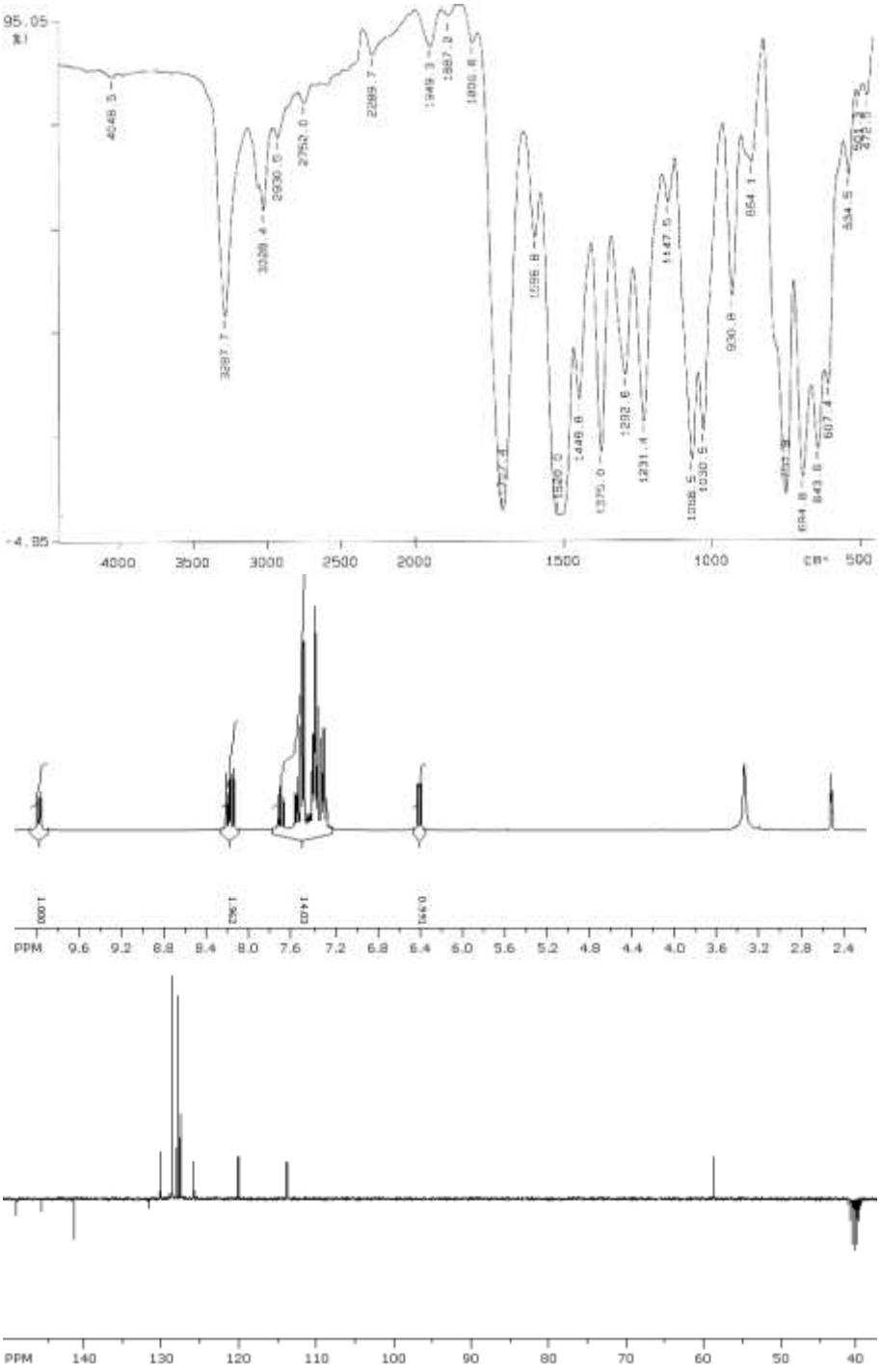

97.95
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2000
1500
1000
cm⁻¹
500

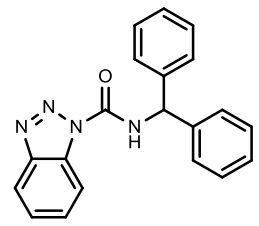

PPM
8.8 8.4 8.0 7.6 7.2 6.8 6.4 6.0 5.6 5.2 4.8 4.4 4.0 3.6 3.2 2.8 2.4 2.0 1.6 1.2 0.8 0.4 0.0


PPM
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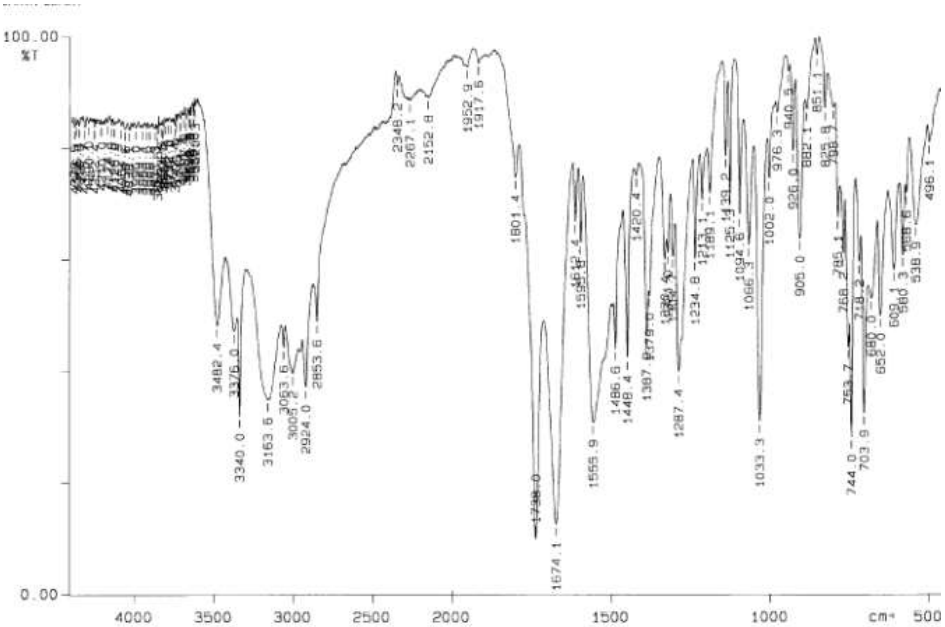

HO
N
O
N
H
N
H
O

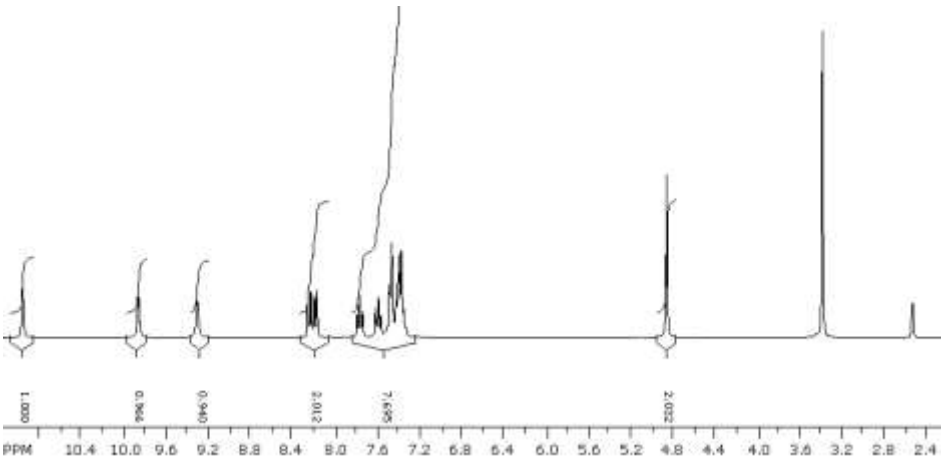
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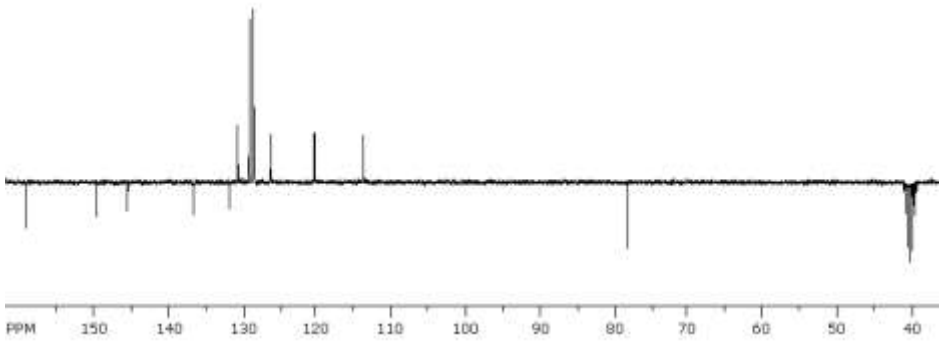


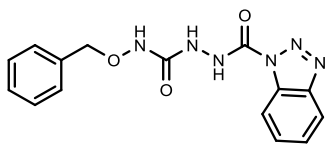


7f

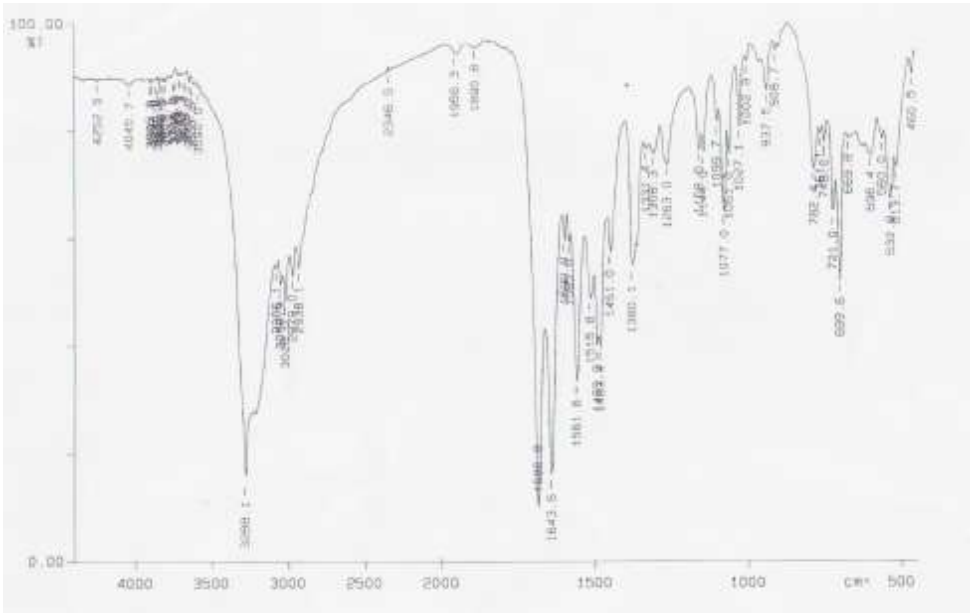

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2000
1500
1000
cm-1
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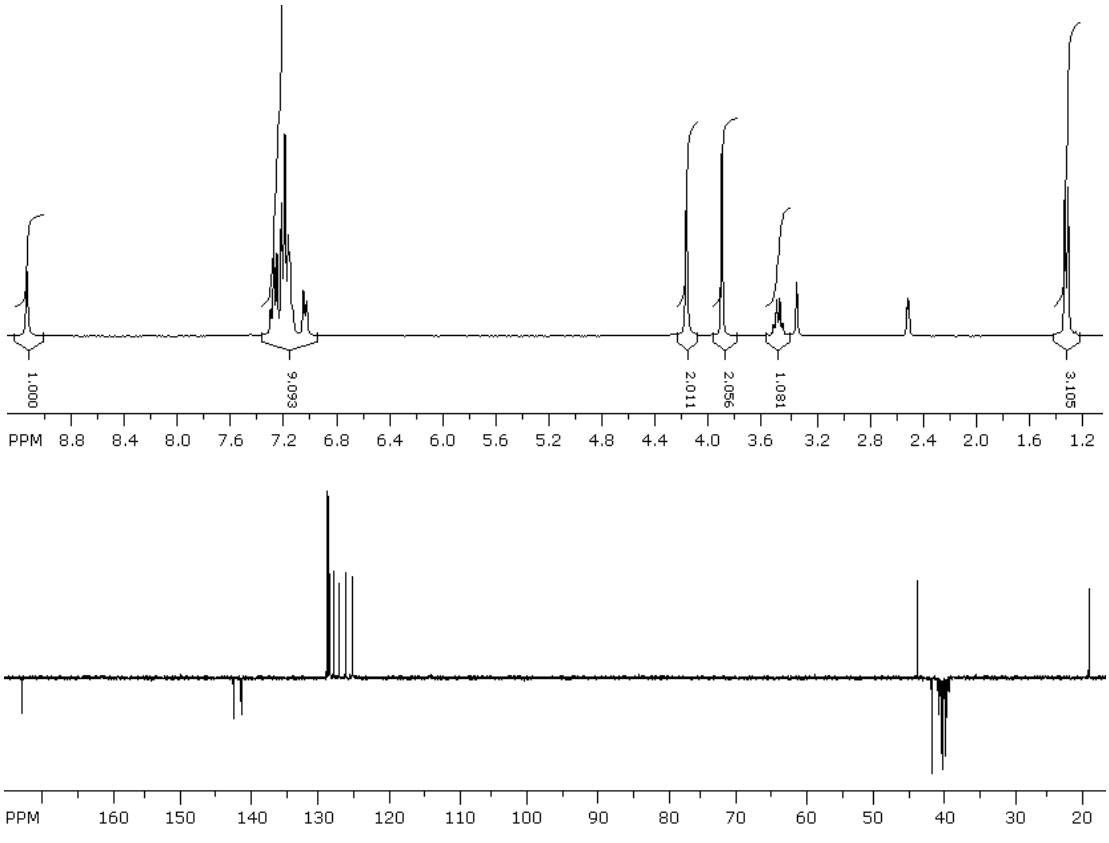

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9.6
9.2
8.8
8.4
8.0
7.6
7.2
6.8
6.4
6.0
5.6
5.2
4.8
4.4
4.0
3.6
3.2
2.8
2.4

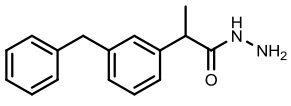

PPM
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80
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60
50
40



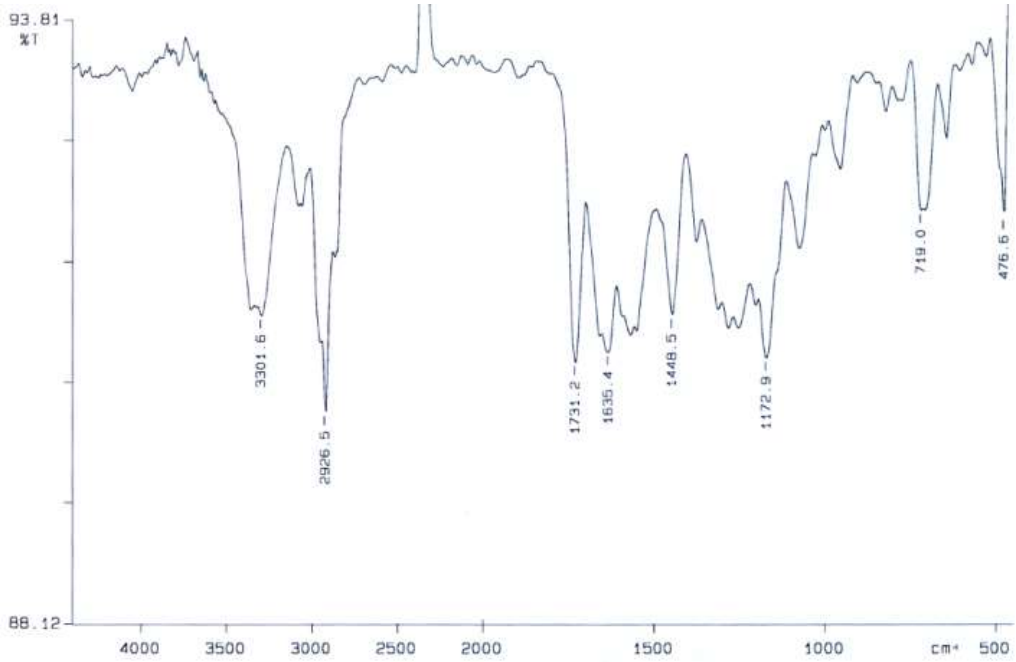
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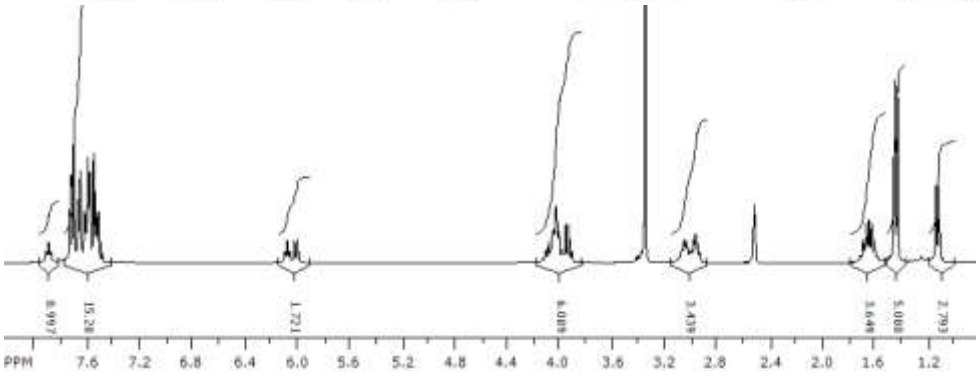

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%T
0.00
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3500
3000
2500
2000
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cm-1
500

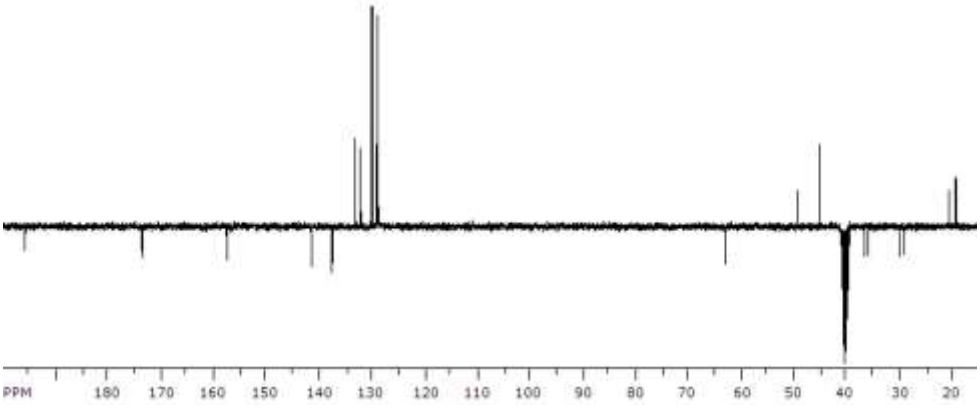

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9.093
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2.056
1.081
3.105
PPM 8.8 8.4 8.0 7.6 7.2 6.8 6.4 6.0 5.6 5.2 4.8 4.4 4.0 3.6 3.2 2.8 2.4 2.0 1.6 1.2
PPM 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20

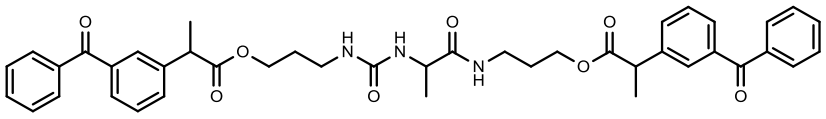

H
N
NH2
O

11c

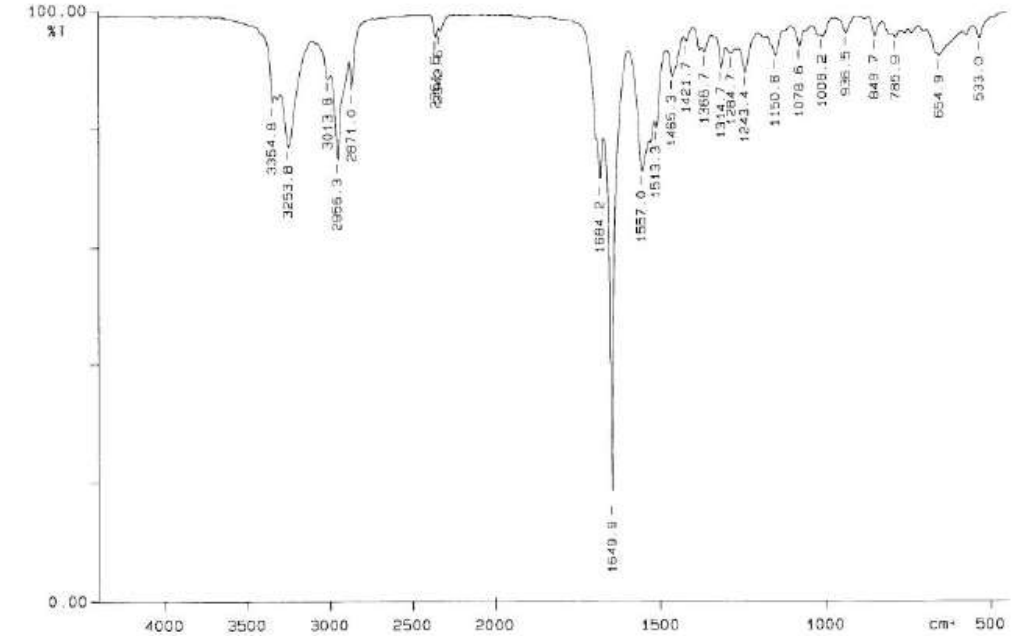

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%T
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2926.5
1731.2
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1172.9
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476.6
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1500
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cm⁻¹
500

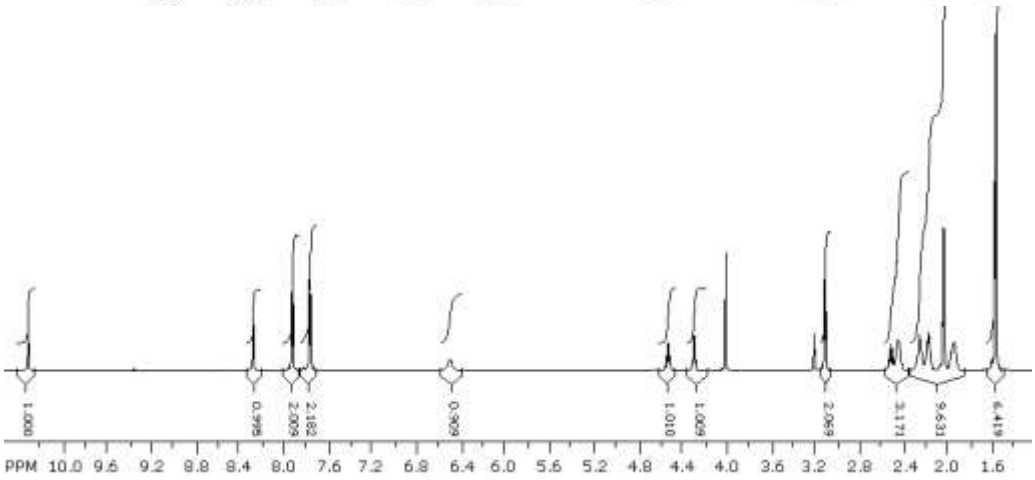

PPM
7.6
7.2
6.8
6.4
6.0
5.6
5.2
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3.6
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2.0
1.6
1.2

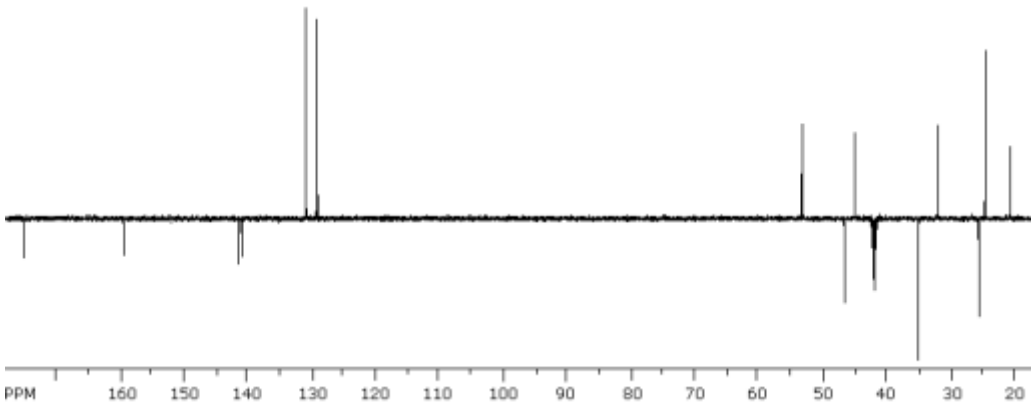

PPM
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160
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140
130
120
110
100
90
80
70
60
50
40
30
20

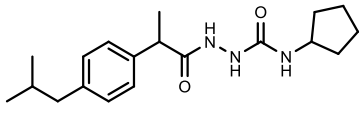


12a

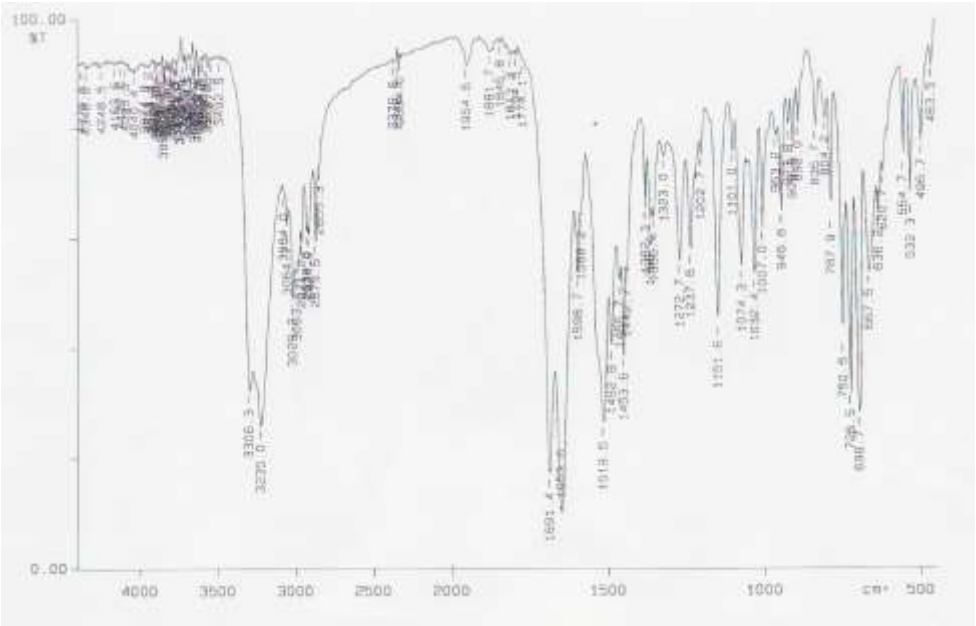

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%T
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3013.8
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2871.0
1684.2
1649.9
1557.0
1513.3
1485.3
1421.7
1366.7
1314.7
1284.7
1243.4
1150.8
1078.6
1008.2
936.5
849.7
785.9
654.9
533.0
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3000
2500
2000
1500
1000
cm-1
500

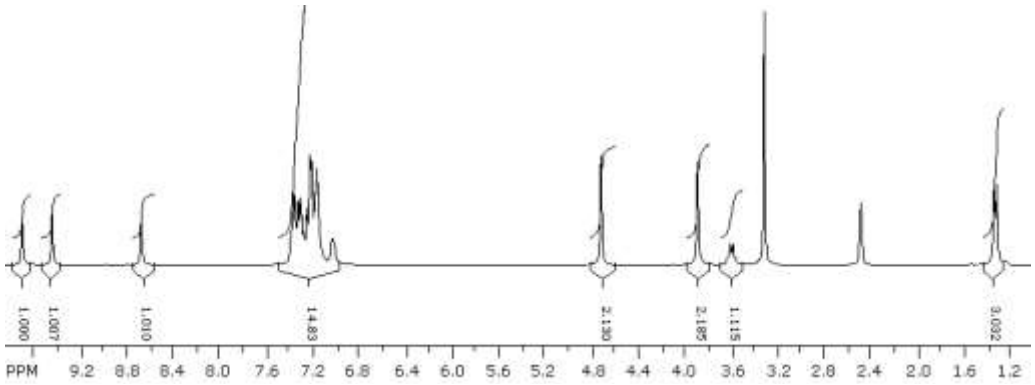

1.000
0.996
2.009
2.182
0.909
1.010
1.000
2.962
3.171
9.631
6.419
PPM 10.0 9.6 9.2 8.8 8.4 8.0 7.6 7.2 6.8 6.4 6.0 5.6 5.2 4.8 4.4 4.0 3.6 3.2 2.8 2.4 2.0 1.6

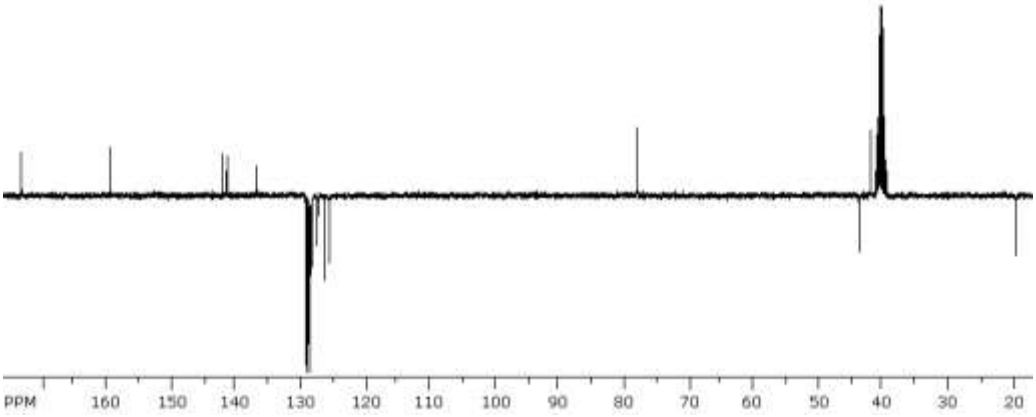

PPM 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20

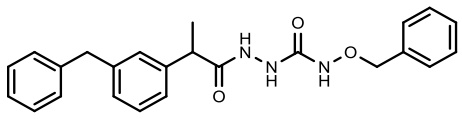


13a

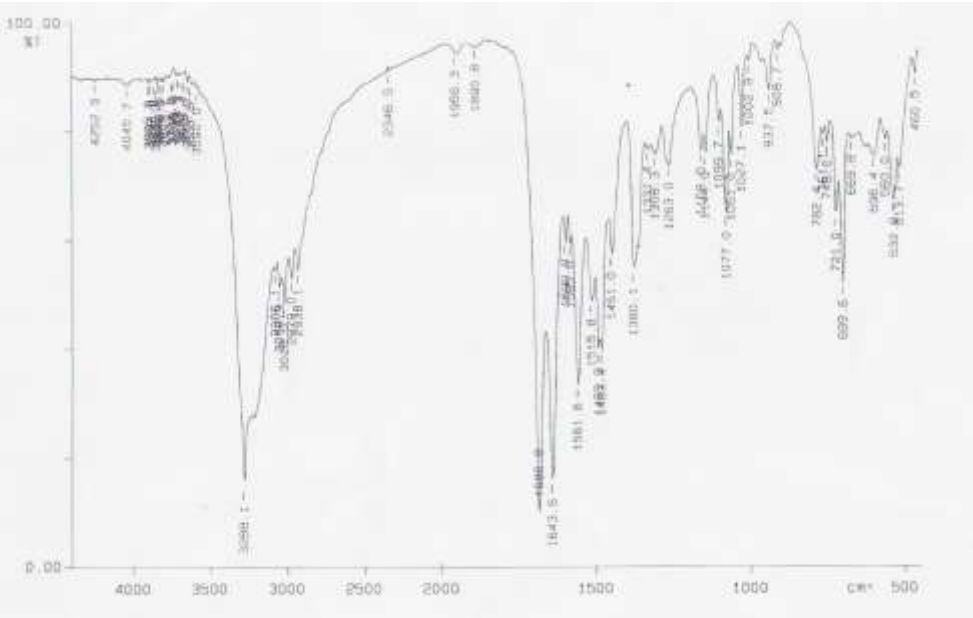


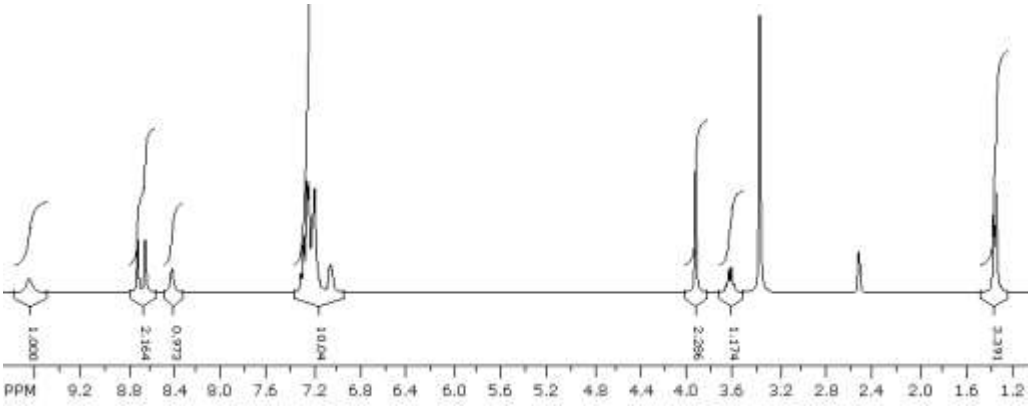


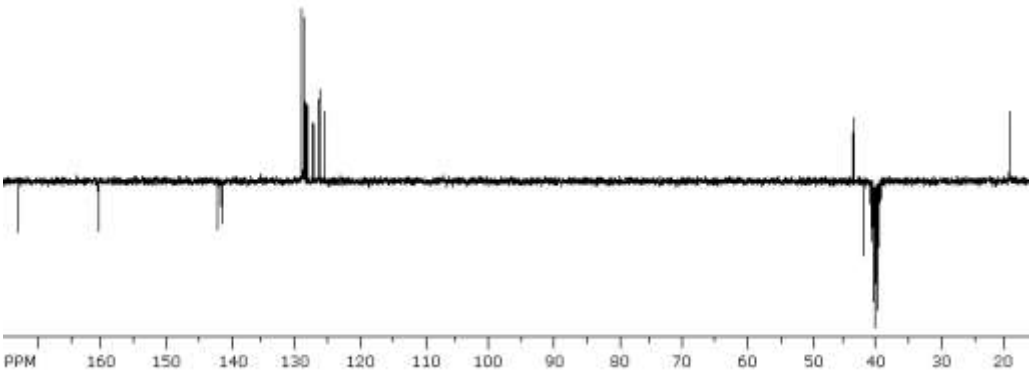


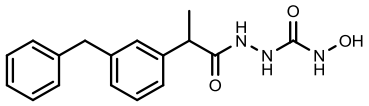


13v

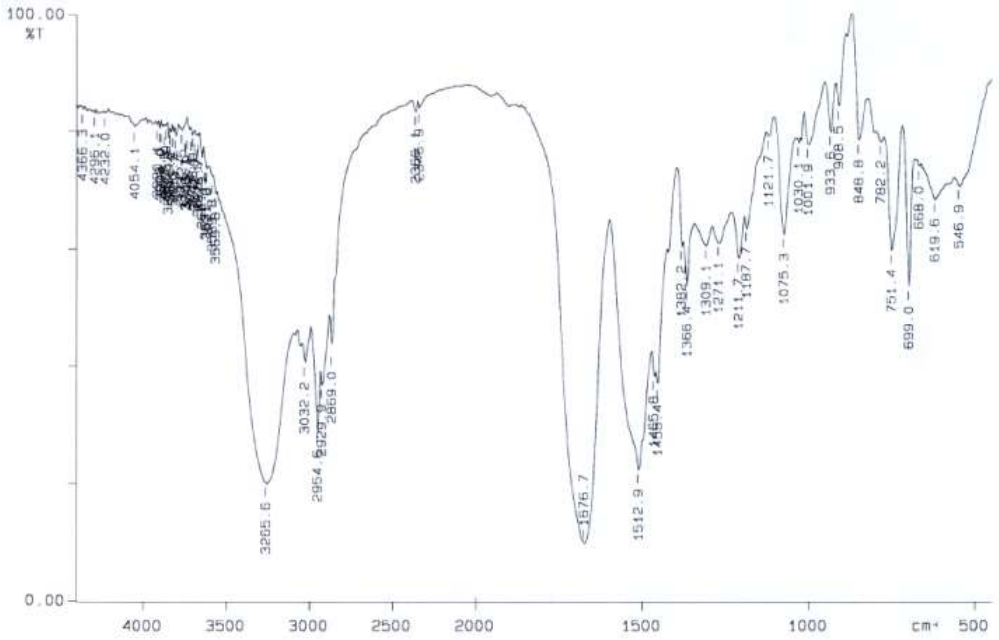


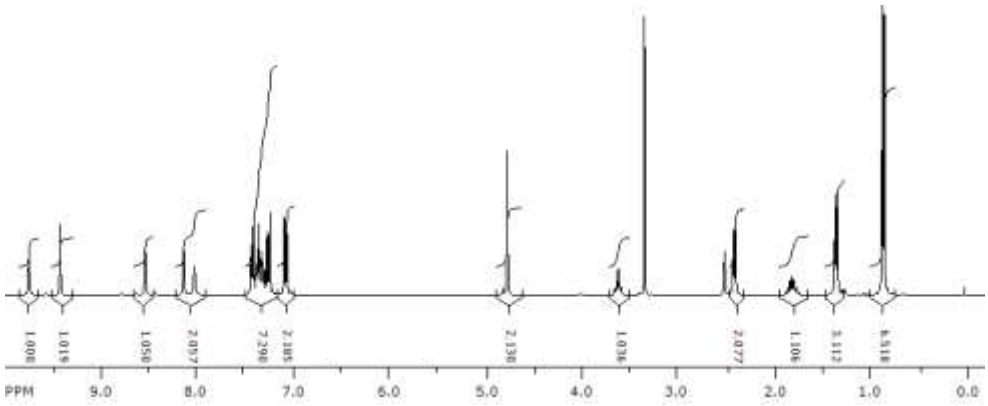

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5.101
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1.174
3.191
PPM
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8.8
8.4
8.0
7.6
7.2
6.8
6.4
6.0
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5.2
4.8
4.4
4.0
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3.2
2.8
2.4
2.0
1.6
1.2

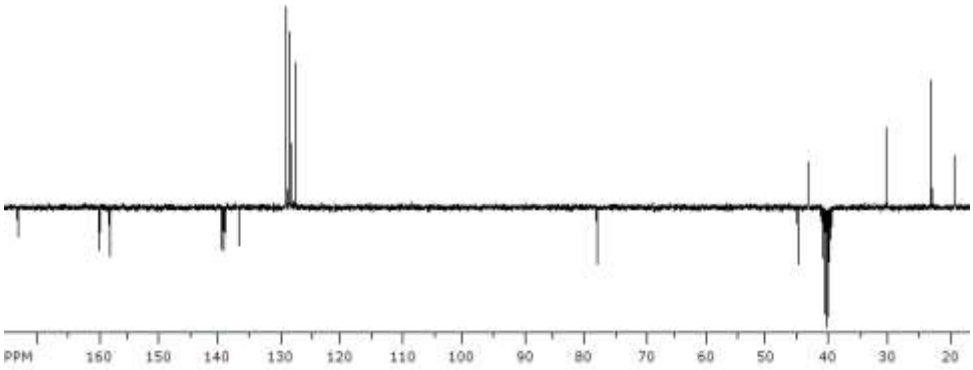

PPM
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20

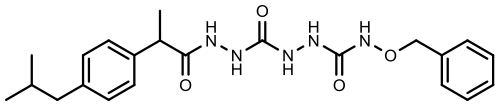

H
N
O
N
H
N
H
O
OH

13y

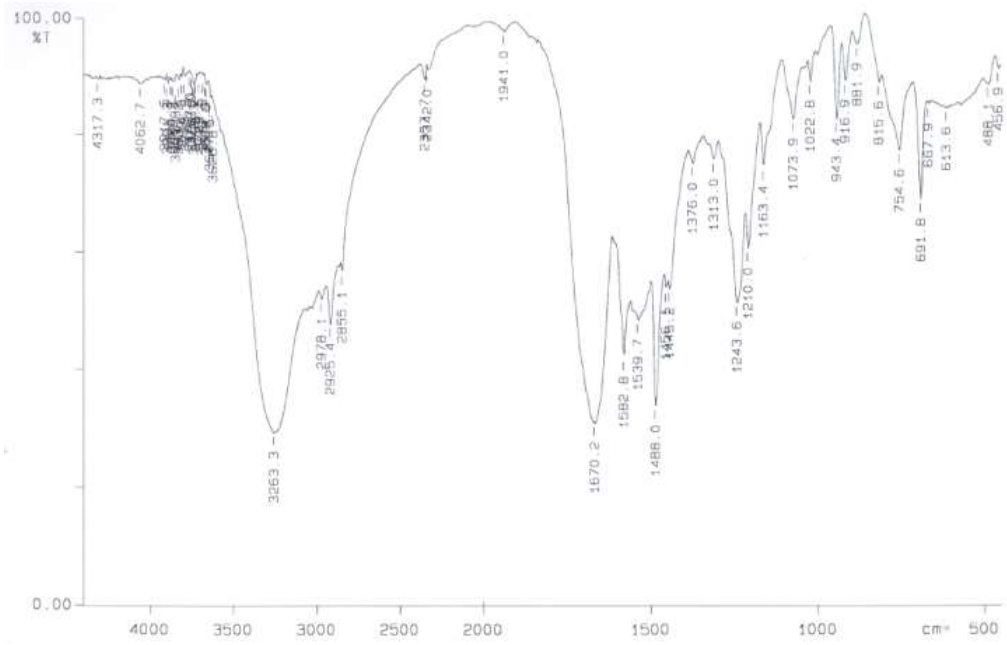

100.00
%T
0.00
4000
3500
3000
2500
2000
1500
1000
cm-
500

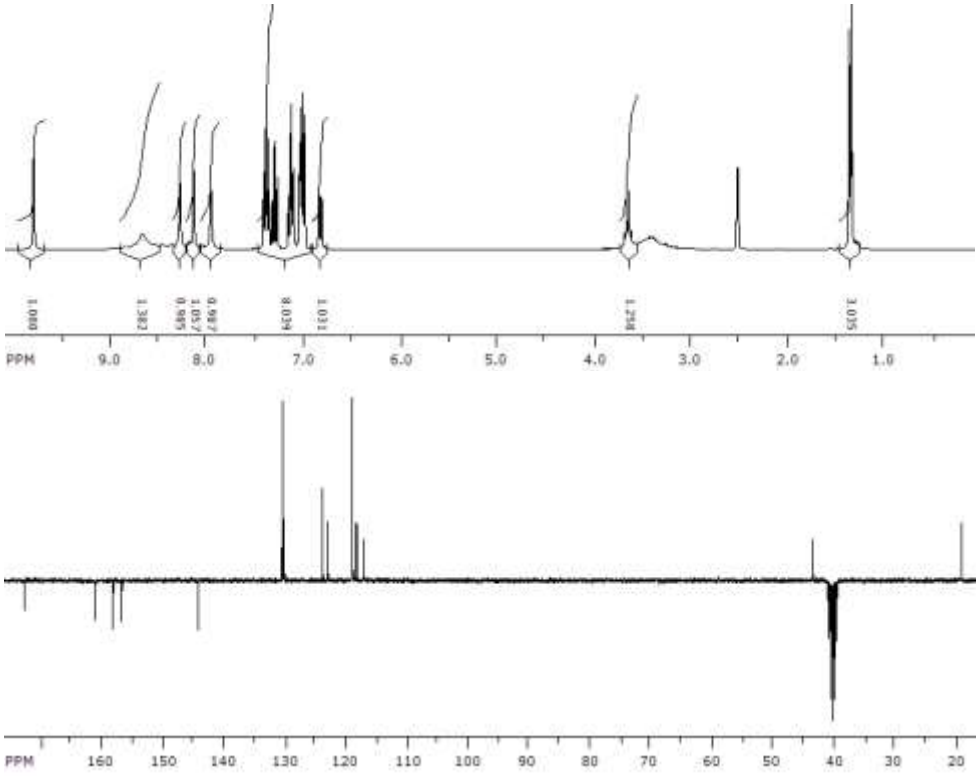

PPM
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8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0

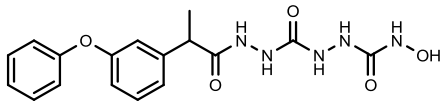

PPM
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20



14a


100.00
%T
0.00
4317.3
4062.7
3263.3
2978.1
2925.4
2855.1
1670.2
1582.8
1539.7
1488.0
1376.0
1313.0
1243.6
1210.0
1163.4
1073.9
1022.8
943.4
916.9
881.9
815.6
754.6
691.8
667.9
613.6
488.1
456.9
4000
3500
3000
2500
2000
1500
1000
cm-1
500


PPM
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
PPM
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20


O
H
N
O
H
N
N
H
O
H
N
OH
O

14e

8. PRILOG B

Prilog sadrži tri znanstvena rada objavljena u časopisima zastupljenim u bazi Current Contents koji obrađuju problematiku iznesenu u ovom doktorskom radu:

Perković I, Butula I, Zorc B, Hock K, Kraljević Pavelić S, Pavelić K, De Clercq E, Balzarini J, Mintas M. Novel Lipophilic Hydroxyurea Derivatives: Synthesis, Cytostatic and Antiviral Activity Evaluations. *Chem Biol Drug Des* **71** (2008) 546–553.

Perković I, Butula I, Rajić Z, Hadjipavlou-Litina D, Pontiki E, Zorc B. Novel Ureidoamides Derived from Amino Acids: Synthesis and Preliminary Biological Screening. *Croat Chem Acta* **83** (2010) 151−161.

Zovko Končić M, Barbarić M, **Perković I**, Zorc B. Antiradical, chelating and antioxidant activities of hydroxamic acids and hydroxyureas. *Molecules* **16** (2011) 6232−6242.

TEMELJNA DOKUMENTACIJSKA KARTICA

Sveučilište u Zagrebu **Doktorski rad**
Farmaceutsko-biokemijski fakultet
Zavod za farmaceutsku kemiju
A. Kovačića 1, 10000 Zagreb, Hrvatska

SINTEZA, KARAKTERIZACIJA I BIOLOŠKO DJELOVANJE KARBAMIDNIH, SEMIKARBAZIDNIH, KARBAZIDNIH I ESTERSKIH DERIVATA AMINOKISELINA I NESTEROIDNIH PROTUUPALNIH LIJEKOVA

Ivana Perković

SAŽETAK

U okviru ovog doktorskog rada, koji je nastavak istraživanja o mogućnostima korištenja benzotriazola u sintetskoj kemiji na Zavodu za Farmaceutsku kemiju Farmaceutsko-biokemijskog fakulteta u Zagrebu, sintetizirano je nekoliko serija spojeva. Polazni spoj u sintezi svih prekursora bio je klorid 1-benzotriazolkarboksilne kiseline (BtcCl) koji daje vrlo reaktivne derivate. BtcCl vrlo lako reagira s nukleofilima dajući cijeli niz reaktivnih spojeva iz kojih je do sada pripravljen velik broj organskih spojeva. Derivati aminokiselina ureidoamidi **4a**-**o** pripravljeni su reakcijom klorida *N*-(1-benzotriazolkarbonil)aminokiselina **3** i odgovarajućih aminoalkohola, a *N*- i *O*- supstituirane hidroksiuree **6a**-**l** iz amida *N*-(1-benzotriazolkarbonil)aminokiselina i odgovarajućih hidroksilamina. Derivati nesteroidnih protuupalnih lijekova (NSAID, ibuprofena, fenoprofena i ketoprofena) sintetizirani su iz NSAID hidrazida **11** koji su dobiveni aminolizom NSAID benzotriazolida **10** i hidrazina. Hidrazidi **11** su u reakciji s 1-karbamoilbenzotriazolima (aktivne uree, **7**) dali 1-acilsemikarbazidne derivate NSAID **13a**-**v**, a u reakciji s 1-(1-benzotriazolkarbonil)-4-benziloksisemikarbazidom (**9**) 1-acil-5-benziloksikarbamoilkarbazide **14a**-**c**. Reakcije u talini uz TEA bitno su skratile vrijeme reakcije. 4-Hidroksisemikarbazidni derivati NSAID **13w**-**y** i **14d**-**f** pripravljeni su katalitičkim hidrogeniranjem *O*-benzilnih derivata **13t**-**v** i **14a**-**c** uz Pd/C. Iz benzotriazolida ketoprofena (**10d**) i odgovarajućih ureidoamida (**4b**,**i**,**j**,**m**) pripravljeni su esteri s dvije molekule ketoprofena kovalentno povezane ureidoamidima u jednu molekulu („dvojni lijekovi“).

Svi sintetizirani spojevi karakterizirani su uobičajenim analitičkim postupcima i spektroskopskim metodama te biološki ispitani *in vitro* na citostatsko, antivirusno, antimikrobno i antioksidativno djelovanje. Nadalje, ispitana je inhibicija lipooksigenaze i lipidne perkosidacije. Najbolje antitumorsko djelovanje pokazao je spoj **6l**, antivirusno **13a**,**g**,**m**, a antiooksidativno spoj **9**. Najjači inhibitor lipooksigenaze bio je spoj **13s**, a lipidne peroksidacije **13c**,**l**,**t**. Uočeno je ponavljanje benzhidrilnog supstituenta u spojevima s istaknutom aktivnosti, što je dobar pokazatelj za daljnje usmjeravanje istraživanja.

Rad je pohranjen u Centralnoj knjižnici Farmaceutsko-biokemijskog fakulteta Sveučilišta u Zagrebu.

Rad sadrži: 183 stranica, 12 slika, 30 shema, 31 tablica i 128 literaturnih navoda. Izvornik je na hrvatskom jeziku.

Ključne riječi: aminokiselina/hidroksiurea/nesteroidni protuupalni lijek/semikarbazid/ester/sinteza/biološko djelovanje

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University of Zagreb **Ph.D. Thesis**
Faculty of Pharmacy and Biochemistry
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SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF CARBAMIDE, SEMICARBAZIDE, CARBAZIDE AND ESTER DERIVATIVES OF AMINO ACIDS AND NONSTEROIDAL ANTIINFLAMMATORY DRUGS

Ivana Perković

SUMMARY

The research described in the thesis is a continuation of the previous work on the use of benzotriazole in the synthetic chemistry which has been carring out at Department of Medicinal Chemistry, Faculty of Pharmacy and Biochemistry in Zagreb for many years. The thesis includes synthesis of several types of compounds. The initial compound for the synthesis of all the intermediates was benzotriazole carboxylic acid chloride (BtcCl), which upon the reaction with nucleophiles gives reactive products, useful in the preparation of various organic compounds. Amino acid derivatives ureidoamides **4a**-**o** were syntehsized by aminolysis of *N*-(1-benzotriazolecarbonyl)amino acid chlorides **3** with corresponding aminoalcohols and *N*- i *O*- substituted hydroxyureas **6a**-**l** were synthesizes from *N*-(1-benzotriazolecarbonyl)amino acid amides **5** and hydroxylamines. Nonsteroidal antiinflammatory drugs (NSAID, ibuprofen, fenoprofen and ketoprofen) derivatives were prepared from NSAID hydrazides **11** which were obtained by aminolysis of NSAID benzotriazolides **10** and hydrazine hydrate. The reaction of hydrazides **11** and 1-benzotriazole carboxylic acid amides **7** gave 1-acylsemicarbazides **13a**-**v**, while with 1-(1-benzotriazolecarbonyl)-4-benzyloxysemicarbazide (**9**) gave 1-acyl-5-benzyloxycarbamoyl-carbazides **14a**-**c**. Reactions in melted state in the presence of TEA significantly reduced the reaction time. 4-Hydroxysemicarbazide compounds **13w**-**y** and **14d**-**f** were prepared by catalytic hydrogenation of *O*-benzylated compounds **13t**-**v** i **14a**-**c** with Pd/C. Ketoprofen benzotriazolide (**10d**) reacted with several ureidoamides (**4b**,**i**,**j**,**m**) and gave esters with two molecules of ketoprofen combined covalently in one („twin drugs“).

The compounds were evaluated for their biological activity *in vitro*: antitumor, antimicrobial, antiviral and antioxidant activity, inhibition of lipoxygenase and linoleic acid lipid peroxidation. The best antitumor activity was exerted by **6l**, antiviral by **13a**,**g**,**m**, antioxidant by compound **9**. The strongest inhibitor of lipoxygenase was **13s**. Compounds **13c**,**l**,**t** showed the highest inhibition of linoleic acid lipid peroxidation. It is worth to note that compounds with the highest biological activity contained benzhydryl substituent.

Thesis is deposited in the Central library of the Faculty of Pharmacy and Biochemistry, University of Zagreb.

Thesis includes: 183 pages, 12 figures, 30 schemes, 31 tables and 128 references. Original is in Croatian language.

Keywords: amino acid/hydroxyurea/nonsteroidal anti-inflammatory drug/semicarbazide/ester/synthesis/ biological activity

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